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PREPARATION AND CHELATE STABILITY OF (ETHYLENEDINITRILO)TETRA-α-PROPIONIC ACID

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ABSTRACT

A method was developed for the synthesis of (ethylenedinitrilo)tetra-α-propionic acid (EDTP) using ethylene diamine and α-chloropropionic acid. The ionization constants of EDTP were determined from pH titrations at 25.0°C and an ionic strength of 0.10. From similar titrations under the same conditions, the metal chelate stability constants for twelve metals with EDTP were determined.

INTRODUCTION

The compound (ethylenedinitrilo)tetraacetic acid (EDTA) has been in industrial use for a number of years as a sequestering agent for metal ions. The versatility and growing importance of EDTA has greatly accelerated interest in other types of amino acids similar to EDTA in structure. The only compounds reported to form metal chelates that have the acetic acid groups replaced by propionic acid groups have these acid groups bonded to the nitrogen through the beta carbon.

This study was made to investigate the synthesis and metal chelates of (ethylenedinitrilo)tetra-α-propionic acid (EDTP). This compound is identical to EDTA except that each of the four acid groups now has a methyl group attached to it. The main interest in this compound is the steric effect of the added methyl groups on the stability of the metal chelates.

EXPERIMENTAL

EDTP preparation. — A mixture of 12 g ethylene diamine (0.2 moles) and 120 g NaOH (3.0 moles) were dissolved in 250 ml water. With rapid stirring, 180 g α-chloropropionic acid (1.65 moles) were added dropwise to the mixture, permitting the temperature to rise no higher than 80°C. The resulting solution was maintained at 80°C for 24 hours. If the pH of the solution fell below 9 during this time, additional NaOH was added. The mixture was cooled and neutralized to pH = 6. Any precipitate (this was found to be the disubstituted product) was filtered. The filtrate was air evaporated to 150 ml. It was divided into six parts of 25 ml each and each part handled separately. To each part was added 10 ml HCl, and NaCl was filtered. The filtrate was added to 10 volumes of ethanol and NaCl again filtered. The filtrate was evaporated, the residue taken up

in 20 ml ethanol, and NaCl was filtered. The filtrate was added to 400 ml acetone and the mixture allowed to set overnight. A fine white powder separated from the solution and was filtered. The solid was the desired EDTP. It was vacuum dried at 50°C for about 5 hours.

Titrations of the product gave an average purity of 98.3 per cent. A nitrogen analysis gave 8.19 per cent (calc. 8.05 per cent). The average yield was 15 per cent.

Reagents. — Solutions of EDTP were prepared approximately 0.002 M in EDTP and 0.110 M in KCl. They were standardized by titration with standard potassium hydroxide. Carbonate-free KOH was prepared by the method of Schwarzenbach and Biedermann (1). The final solution was standardized against HCl that had been standardized gravimetrically. All metal ion solutions were prepared as 0.030 M by weight using reagent grade salts.

Apparatus. — The titration cell was a 150 ml beaker sealed into a glass outer jacket through which water was circulated from a constant temperature bath maintained at 25.0°C. A rubber stopper provided with holes for the electrodes, buret tip, and nitrogen gas inlet and outlet tubes sealed the top of the titration cell. The KOH was added by means of a self-filling 5-ml buret graduated to 0.01 ml. The finely drawn tip of the buret fitted tightly into the rubber stopper and extended below the surface of the solution being titrated. After each addition of base, the solution was mixed by means of a small Teflon-covered bar magnet activated by a magnetic stirrer located under the titration cell. CO_2-free nitrogen, equilibrated with 0.1 M KCl at 25°C, was bubbled through the solution during the titration. A Beckman saturated calomel electrode and a Beckman type E-2 shielded glass electrode (useful over the entire pH range) in conjunction with a Beckman model G pH meter were used to measure pH.

Procedure. — For standardization of the pH meter 4 buffers were prepared according to Bates (2). Their composition and pH at 25°C are as follows: potassium acid tartrate (saturated at 25°C, pH = 3.56); potassium acid phthalate (0.05 M, pH = 4.01); phosphate (0.025 M in each of KH_2PO_4 and Na_2HPO_4, pH = 6.86); and sodium tetraborate (0.01 M, pH = 9.18). The pH meter and electrodes were checked by standardizing the meter against the phosphate buffer at 25.0°C and measuring the pH of the other three buffers. The pH of all three buffers could be reproduced within the reading error of the meter.

In a study of this nature in which concentration constants were desired it became necessary to convert pH from activities to molar concentrations. Bates (3) reports that under very restricted conditions the measured pH can be expected to approach the true activity of the hydrogen ion. These restrictions are that the solutions measured should match the buffers used as reference; namely, aqueous solutions of simple salts with ionic strengths between 0.01 and 0.1. Under these same conditions the Debye-Hückel equation can be used to calculate the hydrogen ion activity coefficient with reasonable accuracy. For this calculation Bates recommends as a value ("ion-size parameter" or "mean distance of approach") of 4 to 6. For an ionic strength of 0.1 at 25°C an average value of log f is -0.10. Thus

$$-\log [H^+] = pH + \log f = pH - 0.10.$$

In some calculations, it was necessary to employ the hydroxide ion concentration. This was obtained by using the ion concentration product for water at 25°C and an ionic strength of 0.10 as given by Harned and Owen (4), $[H^+][OH^-] = 1.60 \times 10^{-14}$.

For the ionization constants of EDTP triplicate titrations were carried out on a 50 ml portion of each of three EDTP solutions diluted with equal volumes of 0.090 M KCl solution. The resulting solutions were about 0.001 M in EDTP and had an ionic strength of 0.10.

For the stability constants of metal chelates with EDTP, duplicate titrations were carried out on a 50 ml portion of each of two EDTP solutions diluted with equal volumes of a 0.030 M solution of a salt of the metal being investigated. The resulting solutions were 0.015 M in the metal ion, 0.001 M in EDTP and had an ionic strength of 0.10.

Experimental results. — The results of one of the titrations of EDTP alone and in the presence of ten of the metals used are given in Figs. 1 and 2. The duplicate titrations (in the case of the free acid, triplicate titrations) were almost identical to those plotted. The titration curves of EDTP in the presence of excess barium and strontium are not plotted as they came quite close to the curve for the free acid.

Calculation of the ionization constants. — An examination of the titration curve for EDTP (curve A, Figs. 1 and 2) shows that the first break in the curve occurs at $\underline{a} = 2$, where $\underline{a}$ is the number of moles of KOH added per mole of EDTP. This means that the first two ionization constants, K_1 and K_2, of EDTP are of the same order of magnitude. Schwarzenbach, Willi and Bach (5) were able to calculate K_1 and K_2 of EDTA by a graphical method. Their method failed to give reasonable results in this case, so that a modified procedure similar to that used by Carini and Martell (6) was derived in which the product of K_1 and K_2 is determined by assuming a ratio of K_1 to K_2. This difficulty arises due to the attempt to calculate two ionization constants whose values are quite close to each other.

The quantities which are known in this system are the total concentration of EDTP (Cs), the hydrogen ion concentration (calculated from the pH measurement), and the fraction of EDTP titrated ($\underline{a}$). The equations available are:

$$K_1 = \frac{[H^+][H_3Y^-]}{[H_4Y]} \tag{1}$$

$$K_2 = \frac{[H^+][H_2Y^{-2}]}{[H_3Y^-]} \tag{2}$$

$$Cs = [H_4Y] + [H_3Y^-] + [H_2Y^{-2}] \tag{3}$$

$$[H^+] = [H_3Y^-] + 2[H_2Y^{-2}] - \underline{a}Cs. \tag{4}$$

Introducing the product and ratio of K_1 and K_2 gives

$$K_1K_2 = K^2 = \frac{[H^+]^2[H_2Y^{-2}]}{[H_4Y]} \qquad (5)$$

$$\frac{K_1}{K_2} = R = \frac{[H_3Y^-]^2}{[H_4Y][H_2Y^{-2}]} \qquad (6)$$

The four equations (3), (4), (5), and (6) contain five unknowns, K, R, $[H_4Y]$, $[H_3Y^-]$, and $[H_2Y^{-2}]$. These can be combined to give one equation with only K and R as the unknowns:

$$X/K^2 + Z\sqrt{R}/K = T \qquad (7)$$

where

$$X = (\underline{a}Cs + [H^+])\ [H^+]^2$$

$$Z = \left[(\underline{a} - 1)Cs + [H^+]\right][H^+]$$

$$T = (2 - \underline{a})Cs - [H^+].$$

Solving equation (7) for K gives

$$K = 2X/\left(-Z\sqrt{R} + \sqrt{Z^2R + 4XT}\right). \qquad (8)$$

For each point on the titration curve from $\underline{a}$ = 0 to $\underline{a}$ = 2, values of X, Z, and T may be calculated. For an assumed value of R, K may be calculated from equation (8). The value of R is chosen such that the values of K calculated from the three titration curves have a minimum average deviation. From the best values of K and R, the first two ionization constants, K_1 and K_2, may be estimated from equations (5) and (6).

Curve A (Figs. 1 and 2) shows distinct breaks at $\underline{a}$ = 2 and a = 3. Thus, only the species H_2Y^{-2} and HY^{-3} need be considered. In this region the determination of K_3 is similar to the determination of K_A for a monobasic weak acid. Thus

$$K_3 = \frac{[H^+][HY^{-3}]}{[H_2Y^{-2}]} = \frac{[H^+]\ (\text{acid titrated})}{(\text{acid untitrated})} = \frac{[H^+]\ (\underline{a} - 2)}{(3 - \underline{a})} \qquad (9)$$

This permits the calculation of K_3 for all values on the titration curve in the range of A = 2.5.

In the basic region of the titration curve from $\underline{a}$ = 3 to $\underline{a}$ = 4, the only species of EDTP that need be considered are $[HY^{-3}]$ and Y^{-4}, hence

$$Cs = [HY^{-3}] + [Y^{-4}] \qquad (10)$$

$$[H^+] + (\underline{a} - 3)Cs = [Y^{-4}] + [OH^-]. \qquad (11)$$

$[H^+]$ may be neglected in this basic region. Combining equations (10)

and (11) with the equation for K_4 gives

$$K_4 = \frac{[H^+][Y^{-4}]}{[HY^{-3}]} = \frac{[H^+]\left[(\underline{a} - 3)Cs - [OH^-]\right]}{(4 - \underline{a})Cs + [OH^-]} \quad (12)$$

Equation (12) permits the calculation of K_4 for each point on the titration curve beyond $\underline{a} = 3$.

Calculation of the chelate stability constants. — The titration curves in Figs. 1 and 2 show that in the presence of excess metal ions EDTP behaves as a stronger acid, especially during the last part of the titration. This increased acidity is used to calculate the stability constants of the metal complexes. In these titrations the following equations can be used:

$$Cs = [H_4Y] + [H_3Y^-] + [H_2Y^{-2}] + [HY^{-3}] + [Y^{-4}] + [MY^{-2}] \quad (13)$$

$$\underline{a}Cs + [H^+] = [H_3Y^-] + 2[H_2Y^{-2}] + 3[HY^{-3}] + 4[Y^{-4}] + 4[MY^{-2}] + [OH^-]. \quad (14)$$

There are five other equations available; namely, the four equations defining the ionization constants of the acid and the equation defining the stability constant of the metal chelate, $K_m = [MY^{-2}]/([M^{+2}]\,[Y^{-4}])$. Combining these equations to eliminate the various species of EDTP, one equation is obtained containing K_m as the only unknown:

$$K_m[M^{+2}](2Cs - b) = b\left(\frac{[H^+]^4}{\overline{K}_1} + \frac{[H^+]^3}{\overline{K}_2} + \frac{[H^+]^2}{\overline{K}_3} + \frac{[H]}{K_4} + 1\right) + Cs\left(\frac{2[H^+]^4}{\overline{K}_1} + \frac{[H^+]^3}{\overline{K}_2} - \frac{[H^+]}{K_4} - 2\right). \quad (15)$$

where $\underline{b} = (\underline{a} - 2)Cs + [H^+] - [OH^-]$,

$\overline{K}_1 = K_1K_2K_3K_4$,

$\overline{K}_2 = K_2K_3K_4$,

and $\overline{K}_3 = K_3K_4$.

Equation (15) is rigorous for all parts of the titration curve. It may be simplified in some cases depending on the pH of the solution. For each point on the curve a value of K_m may be calculated. However, the best accuracy is in the portion of the curve that shows the largest change from that of the free acid; namely, in the region between $\underline{a} = 3$ and $\underline{a} = 4$.

DISCUSSION OF RESULTS

EDTP preparation. — The method outlined was the only one found to give the desired compound. The problem of synthesis of EDTP is apparently increased by the steric effects of the propionic acid. In almost all of the preparations some of the disubstituted compound resulted.

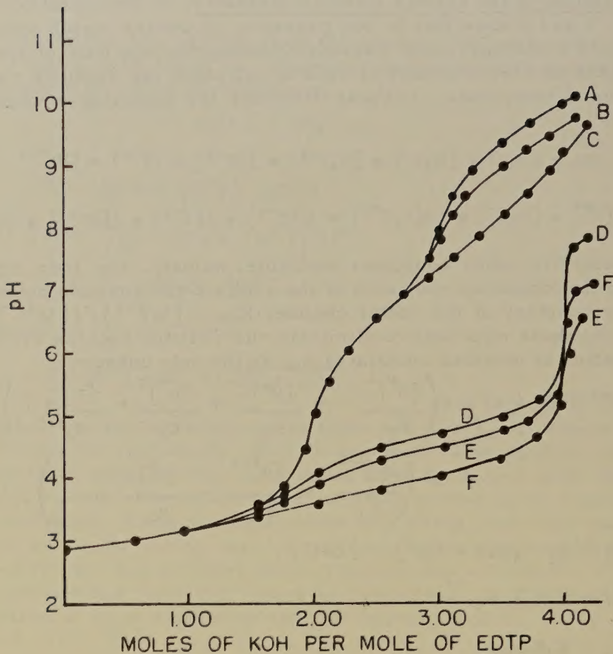

FIGURE I. TITRATION CURVES OF EDTP

CURVE A. EDTP
CURVE B. EDTP + CALCIUM
CURVE C. EDTP + MAGNESIUM
CURVE D. EDTP + CADMIUM
CURVE E. EDTP + MERCURY
CURVE F. EDTP + ZINC

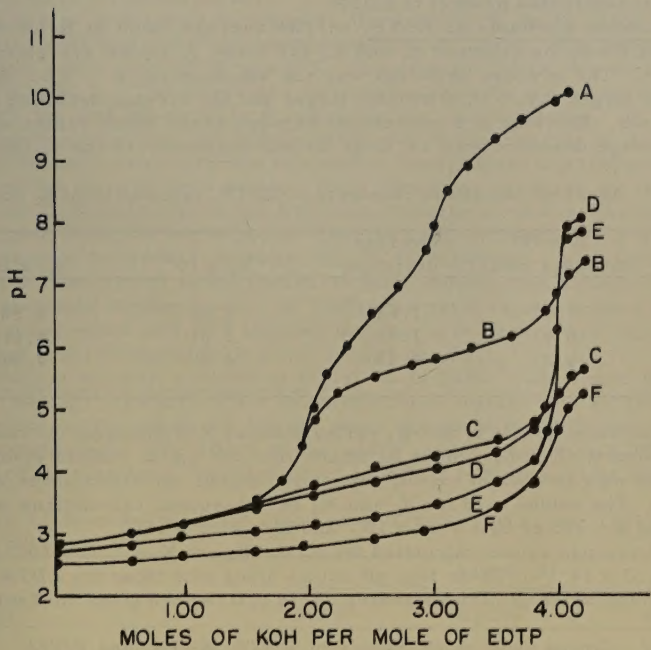

FIGURE 2. TITRATION CURVES OF EDTP

CURVE A. EDTP
CURVE B. EDTP + MANGANESE
CURVE C. EDTP + LEAD
CURVE D. EDTP + COBALT
CURVE E. EDTP + NICKEL
CURVE F. EDTP + COPPER

This always indicated a decrease in the yield and should also indicate the presence of the trisubstituted product. This trisubstituted product must have been eliminated as the hydrochloride in the preparation given since the final product in all cases gave almost the correct analysis for the tetrasubstituted product of EDTP.

Ionization constants of EDTP. — The average value of K for each value of R and the values of K_1 and K_2 for these R values are given in Table 1. The average deviation was the smallest for R = 3.5. As R became larger than 4.0, K became larger and the average deviation also increased. No value of R was chosen between these three values since the average deviation was as large as the difference in the K values.

Table 1. Apparent ionization constants of EDTP, calculation of K_1 and K_2

R	$K \times 10^3$ (average value)	Average deviation	$K_1 \times 10^2$	$K_2 \times 10^3$
2.667	7.82	1.68	1.27	4.80
3.5	8.63	1.38	1.61	4.61
4.0	9.27	1.58	1.85	4.64

It can be seen, however, that K_2 varies little as R is changed, the change being almost all in K_1. Since K_1 enters into the metal chelate stability constant only to a slight extent, this error should not affect these constants. The values used for K_1 and K_2 in subsequent calculations were those of R = 3.5 or $K_1 = 1.61 \times 10^{-2}$ and $K_2 = 4.61 \times 10^{-3}$.

The average values calculated for K_3 and K_4 are $K_3 = 3.72 \times 10^{-7}$ and $K_4 = 5.37 \times 10^{-10}$. These four pK values along with those for EDTA and 1, 2-cyclohexylenedinitrilotetraacetic acid (CDTA) are given in Table 2.

Table 2. Comparison of pK values of EDTP, EDTA, and CDTA

	EDTP[a]	EDTA[b]	CDTA[c]
pK_1	1.89	2.00	2.43
pK_2	2.32	2.67	3.52
pK_3	6.43	6.16	6.12
pK_4	9.27	10.26	11.70

[a]This work. Ionic strength = 0.10; temperature = 25.0°C.

[b]G. Schwarzenbach and H. Ackermann, Helv. Chim. Acta. 30:1798 (1947). Ionic strength = 0.10; temperature = 20.0°C.

[c]G. Schwarzenbach and H. Ackermann, Helv. Chim. Acta. 32:1682 1949). Ionic strength = 0.10; temperature = 20.0°C.

The constants for these latter two chelating agents were determined at 20.0°C but nonetheless provide an interesting comparison. It can be seen that EDTP is the strongest of the three acids except in the case of the third ionization. EDTP would be expected to be the strongest acid

especially for the third and fourth ionizations as the steric effects would reduce the possibility of hydrogen bonding between two carboxyl groups and would also hinder the formation of the amine salt or zwitterion. This reasoning is violated in the case of the third ionization but is upheld by the other three ionizations.

It is also interesting to compare the ratio of K_1 to K_2 since in the present work this ratio was used to calculate K_1 and K_2. The ratio giving the best results for EDTP was 3.5. The ratio for EDTA is 4.67 and for CDTA is 12.3. The closer this ratio approaches the statistical value of 2.667, the less effect the first ionization has on the second. In CDTA the amine groups are in a fixed position due to the rigidity of the ring. Thus the acid groups are kept in a rather fixed position and they probably are rather close to each other. Their closeness is shown by the fact that the first ionization is much stronger than the second, or the negative charge obtained on the molecule from the first ionization has a greater influence on the second ionization due to its nearness. For EDTA the ratio is much closer to the statistical value showing that the acid groups are generally farther apart. In EDTP the ratio is still closer to the statistical value of 2.667 showing that the acid groups are kept still farther apart by the addition of the methyl groups.

Stability of metal chelates of EDTP. — In Table 3 are listed the log K_m (stability constant) values calculated for 12 metals with EDTP. Also included are the constants for the same metals with EDTA and CDTA for comparison. The effect of the added methyl groups in EDTP can immediately be seen. The large over-all effect is to reduce the chelating

Table 3. Stability of metal chelates of EDTP, EDTA, and CDTA

	Log K_m		
Metal	EDTP[a]	EDTA[b]	CDTA[b]
Magnesium	3.03	8.69	10.32
Calcium	2.01	10.96	12.08
Strontium	1.14	8.63	c
Barium	1.22	7.76	7.99
Zinc	9.68	16.50	16.67
Cadmium	8.33	16.46	19.32
Mercury	8.72	21.8	24.3
Manganese	6.06	14.04	16.78
Nickel	11.0	18.62	c
Cobalt	9.70	16.31	18.92
Copper	13.0	18.80	21.30
Lead	9.31	18.04	19.68

[a]This work, Ionic Strength = 0.10; temperature = 25.0°C.

[b]G. Schwarzenbach, R. Gut, and G. Anderegg, Helv. Chim. Acta, 37, 937 (1954). Ionic strength = 0.10; temperature = 20.0°C.

[c]Data not given.

ability of EDTP greatly. The explanation is the steric effect offered by the methyl groups to the metal being chelated. There is also secondary effect due to the addition of the methyl groups and that is one of interest, namely the relatively stronger complex with smaller ions. This is shown in the case of megnesium and calcium. With EDTA calcium forms the stronger chelate by two orders of magnitude. However, with EDTP the smaller ion, magnesium, forms the stronger complex by a factor of ten. Again this effect is noted with zinc and cadmium whose stability constants with EDTA are about the same. With EDTP the smaller ion, zinc, forms the stronger complex by more than a factor of ten. It appears that this steric effect breaks down in the case of barium and strontium since with EDTP the larger ion forms the stronger chelate while the reverse is true with EDTA. This may be explained in part by the difficulty in the calculations of such low magnitude stability constants. The difference between the titration curve of barium and strontium with EDTP and that for EDTP only is slight, thus causing relatively large errors.

It should be pointed out that in the case of the constants of nickel, zinc, cobalt, and copper, there is a continual drift of the constants during the course of the titration. For this reason, only points in the last fourth of the titration were used for calculation. Even here the shift is quite significant and becomes more so the stronger the complex. This may be due to two effects. The first one is that the method employed is beginning to give erratic results since it is being used at its upper limit. This effect is pointed out by the fact that lowering the pH value 0.01 units for a point in the copper titration doubles the value of the stability constant at that point. The only way to correct this would be to change to another technique. The second effect is that the stronger the complex the more acid is the solution. This may enhance the formation of acid-metal chelates of the form MHY^- or even MH_2Y. These species were neglected in the present work since for most cases the effect would be small.

The effect of chloride on the stability constants of mercury and lead (and perhaps cadmium) is rather hard to consider. Equation (15) used to calculate the metal chelate stability constants contains the free metal ion concentration only once and this appears in the denominator of the expression of K_m. In the case of mercury, for instance, the free metal ion would not be the value assumed since almost all of the mercury was not free but complexed by chloride. If one assumes the mercuric chloride is one per cent ionized, the free metal ion concentration would be one hundredth the value assumed in the original calculations. This would increase the value of the stability constant by two orders of magnitude or $\log K_m = 10.72$. This value would certainly be a better estimate of the EDTP-mercury complex than the value listed.

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THE SUCCINOXIDASE SYSTEM IN MYROTHECIUM VERRUCARIA[1,2]

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Cytoplasmic particles which contain the entire complement of Krebs cycle enzymes and co-factors have been isolated from both plant and animal tissues. The properties of many of the individual enzyme systems have been studied extensively with particles of animal origin and to a considerable though lesser extent with those of higher plant origin. Knowledge of the occurrence of particulate respiratory enzymes and of their properties in the filamentous fungi is much less extensive. The present experiments on the succinoxidase system are part of a study of mechanism of aerobic respiration and of the action of fungicides in the mycelium of Myrothecium verrucaria (Alb. and Schw.) Dit. ex. Fr. Reports have appeared on the general nature of the respiration of this organism (5) and on the occurrence and properties of cytochrome oxidase (6), ascorbic oxidase (13), and malic dehydrogenase (8).

METHODS AND MATERIALS

Mycelial suspensions of M. verrucaria QM 460 were produced in shake culture as described by Walker (23). Under our conditions 24-hour cultures at 30°C gave mycelium of about maximum Q_{O_2} (60-75) in the form of rather loose, flexible clumps rather than distinctly spherical pellets. Particle extraction was carried out as follows. The culture medium was removed by suction filtration and the mycelium washed three times with water. The mycelial mat of 10-12 g wet weight then was blended for 30 seconds in a Waring Blendor containing 150 ml of cold 0.01 M pH 7 KH_2PO_4-Na_2HPO_4 buffer and centrifuged to remove most of the liquid. The mycelial pads were ground in a 60-ml Potter-Elvehjem homogenizer in an ice bath with an equal weight of 100-mesh pyrex glass and 2-4 ml of the pH 7 buffer. The homogenate was diluted to about 50 ml with the same buffer and centrifuged at 0-5°C for 5 minutes at 600 x g to remove glass, cellular debris, and unground mycelial fragments. The residue then was subjected to a second grinding procedure like the first. The two supernatant fractions, which were free of intact cells by microscopic

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[2]Journal Paper No. J-3234 of the Iowa Agricultural Experiment Station, Ames, Iowa. Project 1258. These studies were aided by a contract between the Office of Naval Research, Department of the Navy, and Iowa State College, NR-103-198.

[3]Present address: Crops Research Division, Agricultural Research Service, U.S. Department of Agriculture, Beltsville, Maryland.

inspection, were centrifuged in the cold for 30 minutes at 20,000 x g. The particle fraction sedimented in this way contained all the extracted succinoxidase activity. The supernatant was discarded and the particles were resuspended in 8 ml of cold buffer. On the average, 0.6 ml of such suspensions, containing 0.5-0.8 mg of nitrogen, was added to each 3.0 ml reaction mixture. Flasks were kept chilled prior to the 10-minute temperature equilibration, and oxygen uptake was measured at 30°C in air by standard Warburg procedure.

For respiratory measurements, the mycelium usually was harvested and washed by centrifugation; however, in the tests of malonate and cyanide sensitivity it was harvested and washed by filtration and then blended as described for particle preparation. The assay procedure was essentially that of Darby and Goddard (5) except that a Robbie (19) KCN-KOH center well mixture was used in the cyanide tests.

Special materials, with abbreviations or symbols used hereafter, were from the following sources: adenine, diphosphopyridine nucleotide (DPN), and triphosphopyridine nucleotide (TPN) -- Schwarz; adenosine-5'phosphate (AMP), adenosine diphosphate (ADP), adenosine triphosphate (ATP), uridine triphosphate (UTP), uridine-5'-phosphate (UMP), and coenzyme A(CoA) -- Pabst; cytochrome c -- Sigma; cocarboxylase -- Merck; 2-hydroxy-3-(2-methyloctyl)-1, 4-naphthoquinone (SN5949) -- Abbot; 2, 4-dinitrophenol (DNP) -- Eastman; ethylenediamine tetraacetic acid (EDTA) -- Alrose.

RESULTS

Factors Affecting Oxidative Capacities of Isolated Particles

In preliminary experiments to devise a suitable method for extraction of the succinoxidase system from *M. verrucaria* mycelium a number of procedures based on those of Laties (10) were tried. The principal variables were grinding time, glass-mycelium-fluid proportions, extraction medium (0-0.8 M sucrose and/or 0-0.1 M phosphate), and extent of washing. Many of these particle preparations were tested for ability to oxidize pyruvate, ketoglutarate, fumarate, citrate, and malate as well as succinate, but only the latter was oxidized and at very low rates. When particle washing was omitted the rate of succinate oxidation was increased and some ketoglutarate oxidation was observed. Significant rates of oxidation of the other Krebs cycle intermediates occurred on addition of a mixture of cofactors including ATP, cytochrome c, CoA, cocarboxylase, Mg, TPN, and DPN. However, even with these additions the oxidative capacities of particle preparations were quite variable, and the variations could not be correlated with changes in extraction procedure. In the presence of small amounts of malate, pyruvate was oxidized with Q_{O_2} (N) values ranging from 47 to 340. Ketoglutarate values varied from 82-275. The values for malate and fumarate ranged from 0 to as high as 300 but were seldom over 50. Under the same conditions, unwashed particles by various extraction procedures and with the above cofactors, succinate Q values were from 130 to 530. In contrast to the other substrates, oxidation of succinate was consistently faster with only ATP and cytochrome c added than with the more complete cofactor mixture. A typical experiment of this kind is shown in

Table I. Furthermore, there appeared to be somewhat less variation in succinoxidase activity among particle preparations when they were tested without the additional cofactors. These observations suggested that products of concatenary reactions interfered in succinate oxidation.

In subsequent work on the succinoxidase system the extraction procedure in the methods section was used unless otherwise specified. The *M. verrucaria* particles were relatively insensitive to phosphate concentration; however, in most trials they gave slightly higher rates at 0.01 M than at higher or lower concentrations, and this level was used both in extraction and in the reaction mixture. Further evidence of variation among particle preparations appeared in their response to succinate concentration. Lineweaver-Burk (12) plots gave apparent Michaelis constants which ranged from 0.0064 to 0.034 and averaged 0.021 M. Preparations also varied in response to added cytochrome c. A few showed no response, but with the majority stimulation averaged about 100%. Maximal stimulation in all cases was reached with 1 mg per flask (ca. 1.1×10^{-5} M). When saturated with substrate and cytochrome c, the pH of maximum activity for all preparations was about 7.

Nature of Adenylate Stimulation

With the succinoxidase system apparently saturated with substrate and cytochrome c the next step toward optimum reaction conditions appeared to lie in investigating the action of ATP. Like several higher plant particle preparations (11, 15, 17) the fungus succinoxidase was stimulated by added ATP. The increase in rate was usually from 50 to 100% and occasionally as much as 200 %. Furthermore, rates were constant for longer periods. Maximum effect with ATP was at 0.001 M. ADP was equally active in all trials, but particle preparations varied in their response to AMP. With some preparations, AMP was nearly as active as ATP while with others it caused no increase in rate. Table II summarizes results of a typical experiment with the former type of preparation. Some stimulation was always observed with UTP but never with UMP, and adenine and pyrophosphate were inactive. In no case could the increased oxygen uptake be attributed to further oxidation of fumarate (cf. Table I).

Since higher plant particles have been activated by Mg as well as ATP (11, 15, 17), a number of the mycelial preparations were tested for a Mg effect both with and without added adenylate. Although succinoxidase activity was stimulated, the increase in rate was generally less than 10%. However, an indication of cation participation in the adenylate effect was observed in experiments in which EDTA was added to the reaction mixture. Inhibition of succinate oxidation by 0.01 M EDTA was greater with either AMP or ATP added (av. 50%) than without (av. 15%). With several particle preparations showing pronounced stimulation by AMP (70-100%), it was found that only 0.0005 M EDTA was required to prevent the stimulation. To get more definite evidence of cation participation, particles were prepared in the presence of 0.01 M EDTA by the usual procedure and then washed twice with 0.01 M pH 7 phosphate. Of 9 such preparations, 7 no longer were significantly stimulated by AMP but still showed a definite response to ATP and ADP. This residual adenylate effect was prevented by addition of 0.01 M EDTA to the reaction mixture. Table III includes results of a typical experiment. None of the cations added,

Table I. Effect of cofactors on oxidative capacities of particles.

Substrate (0.043 M)	O_2 uptake (µl/hr. x flask)* ATP + cyto. c	"Complete mixture"
Pyruvate	0	0
Pyruvate + 0.002 M malate	-	69
Citrate	0	83
Ketoglutarate	15	43
Succinate	243	173
Fumarate	0	37
Malate	7	12

* All flasks contained 0.001 M ATP, 1.1×10^{-5} M cyto. c, 0.01 M pH 7 PO_4, and equal volumes of particle suspension. "Complete mixture" also included 0.05 mg TPN, 1 mg DPN, 0.03 mg CoA, 0.05 mg cocarboxylase, and 0.001 M $MgCl_2$.

Table II. Effect of adenosine and uridine nucleotides and derivatives on succinoxidase activity.

Treatment* (0.001 M)	O_2 uptake (µl/hr X flask)	Treatment* (0.001 M)	O_2 uptake (µl/hr X flask)
Control	121	UTP	220
Pyrophosphate	116	AMP	356
Adenine	120	ADP	392
UMP	120	ATP	393

* All flasks contained 0.043 M succinate, 0.01 pH 7 phosphate, and 1.1×10^{-5} M cyto. c.

Table III. Effect of cations on succinoxidase activity of particles prepared in EDTA.

Cation treatment*	O_2 uptake (μl/hr X flask) Control	0.001 M AMP	0.001 M ADP	0.001 M ATP
Control	140	155	200	210
0.01 M EDTA	-	-	-	153
0.001 M $MgCl_2$	150	166	204	204
0.001 M $MnCl_2$	142	159	206	204
Control	178	175	-	206
0.001 M $FeCl_3$	176	182	-	198
0.001 M $CoCl_2$	44	110	-	190
0.00003 M $CuSO_4$	80	84	-	106

* All flasks contained 0.043 succinate, 0.01 M pH 7 phosphate, and 1.1×10^{-5} M cyto. c.

Table IV. Effect of ATP on inhibition of succinoxidase activity by dicarboxylic acids.*

Inhibitor	No. of Expts.	Average percentage inhibition Control	0.001 M ATP
0.0007 M Malonate	4	47	46
0.007 M Aspartate	4	12	9
0.007 M Fumarate	3	34	14
0.007 M Malate	3	44	8
0.007 M Oxalacetate	3	93	84
0.00075 M Oxalacetate	4	92	56
0.007 M Pyruvate	3	-3	1

* All flasks contained 0.043 M succinate, 0.01 M pH 7 phosphate, and 1.1×10^{-5} M cyto. c.

Table V. Removal of oxalacetate inhibition of succinoxidase activity by ATP and glutamate.*

Succinate (M)	Oxalacetate (M)	O_2 uptake (μl/hr x flask)		
		Control	ATP (0.001 M)	Glutamate (0.04 M)
0	0	-	-	0
0.043	0	228	520	616
0.043	0.00001	211	484	628
0.043	0.00004	166	460	626
0.043	0.0001	88	436	596

* All flasks contained 0.01 M pH 7 phosphate and 1.1×10^{-5}M cyto. c.

Table VI. Effect of cyanide on succinoxidase activity and on mycelial respiration.

Cyanide (M)	Per cent Inhibition (aver. and range, 4 expts.)	
	Succinoxidase*	Respiration**
4.6×10^{-6}	11 (4-19)	11 (3-21)
10^{-5}	22 (6-35)	13 (3-24)
4.6×10^{-5}	35 (21-52)	10 (0-27)
10^{-4}	50 (42-62)	17 (0-30)
4.6×10^{-4}	84 (79-88)	65 (37-87)

* 0.043 M succinate, 0.01 M pH 7 PO_4, and 1.1×10^{-5}M cyto. c.
** 0.02 M glucose, 0.001 M $MgSO_4$, and 0.0167 M pH 7 PO_4.

including Zn, Ba, Ca, Al, Mg, Mn, Fe, Co, and Cu, restored AMP stimulation or increased that by ATP. Typical results for 5 of the cations most extensively studied are shown in Table III. The only marked cation effects observed were inhibitions by Co and Cu. With 0.001 M Co, inhibition averaged 66% and was reduced to 34% and 16% by 0.001 M AMP and ATP, respectively. Sensitivity to Cu was greater and was little influenced by addition of adenylates. Although the effect of EDTA indicated that cations influence adenylate stimulation of succinate oxidation by the fungal particles, the evidence is not conclusive and the identity of such cations remains obscure.

In further work on the nature of the ATP stimulation, it was first shown that the effect was not on the oxidation of cytochrome c since no stimulation was observed when ascorbate or hydroquinone replaced succinate as reductant. Furthermore, examination of reciprocal plots (12) indicated that ATP had no effect on the maximum velocity (V_{max}) but appeared to counteract a competitive inhibitor. A typical result is shown in Fig. 1. Malonate and possible reaction products of succinate oxidation then were tested for inhibitory effects in the presence and absence of ATP. As shown in Table IV, ATP did not relieve malonate inhibition but did reduce inhibition obtained in the presence of fumarate, malate, and low concentrations of oxalacetate. The 0.007 M concentrations used in these experiments were the level of products which might be formed in an average 30 minute succinoxidase reaction. Although oxidation of fumarate and malate in the succinoxidase reaction system was only barely detectable manometrically, sufficient oxalacetate could have been formed to explain the observed inhibition of succinate oxidation. The low concentration of oxalacetate required for inhibition is further evidenced in Table V. It seemed likely, therefore, that ATP acted to suppress accumulation of oxalacetate.

Transamination also was investigated as a means of trapping oxalacetate as proposed by Eisenberg (7). L-Glutamate proved to be even more effective than ATP in most trials in stimulating succinate oxidation. A representative experiment is summarized in Table V. Glutamate and ATP stimulations were not additive and the former could completely replace the latter with most particle preparations. Glutamate itself was not oxidized. As in the case of ATP addition, glutamate did not alter the V_{max} but appeared to remove a competitive inhibitor. Experiments like that in Fig. 2 indicated a K_m value of about 0.007 M for succinate and showed that at least 95% of the V_{max} was measured under the following assay conditions: 0.01 M pH 7 phosphate, 0.1 M succinate, 0.04 M glutamate, and 1 mg cytochrome c (ca. 1.1×10^{-5} M). Under these conditions the Q_{O_2} (N) value for eight typical particle preparations ranged from 455-730 and averaged 580. With this assay system good proportionality was observed between particle concentration and oxygen uptake to rates of about 400 microliters per hour (cf. curve 1, Fig. 4).

Attempts to show oxalacetate formation from succinate oxidation by chromatographic detection of aspartate in the glutamate trapping system was unsuccessful because of free aspartate in the particle preparations at about the expected level of oxalacetate accumulation ($1\text{-}2 \times 10^{-5}$ M). However, when 10^{-4} M oxalacetate was added to reaction mixtures containing 5×10^{-4} M glutamate definite formation of aspartate was demonstrated. Furthermore, assay of several particle preparations for trans-

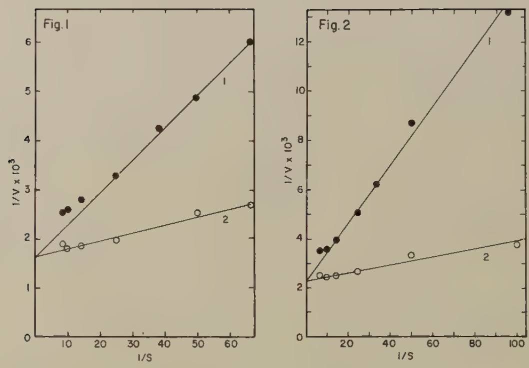

Fig. 1. Test of nature of ATP stimulation by method of Lineweaver and Burk. (V: reaction velocity as O_2 uptake in µl/hr X flask; S: succinate molarity.) Curve 1: no added ATP. Curve 2: 0.001M ATP.

Fig. 2. Test of nature of glutamate stimulation by method of Lineweaver and Burk. (V: reaction velocity as O_2 uptake in µl/hr X flask; S: glutamate molarity.) Curve 1: no added glutamate. Curve 2: 0.04M glutamate.

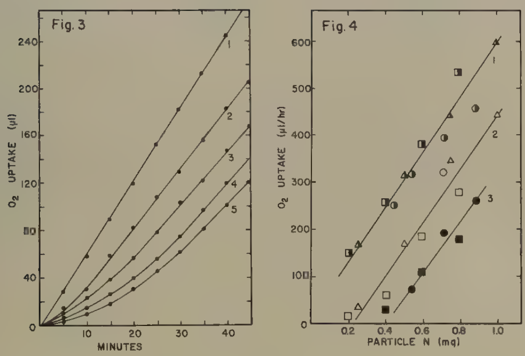

Fig. 3. Decrease in SN5949 inhibition of mycelial respiration with time. Curve 1: no SN5949. Curves 2, 3, 4, and 5: 0.33 X 10^{-6}M, 0.7 X 10^{-6}M, 1.4 X 10^{-6}M, and 2.1 X 10^{-6}M SN5949, resp.

Fig. 4. Relation of SN5949 and particle concentrations on succinoxidase activity of 3 representative particle preparations (prep. A, circles; prep. B, triangles; prep. C, squares). Curve 1: (half-closed symbols) without SN5949. Curve 2: (open symbols) 0.33 X 10^{-6}M SN5949. Curve 3: (closed symbols) 0.55 X 10^{-6}M SN5949.

aminase activity essentially by Karmen's method (9) showed activities of 0.5-1.0 micromoles per minute per ml. This activity would appear to be sufficient to remove oxalacetate formed during succinate oxidation. However, in the case of a few particle preparations glutamate was not fully effective in suppressing inhibition of succinoxidase action. It seems likely that this was due to low transaminase activity since preliminary evidence indicates that most of the transaminase activity of the mycelial homogenates is in the supernatant rather than in the particle fraction and that the activity of the latter can be reduced by resedimentation from buffer.

Effect of Inhibitors on Particles and Mycelium

The action of several inhibitors on succinate oxidation by particles and on oxygen uptake by mycelium was investigated to further characterize the oxidase system and, if possible, to provide evidence of its participation in respiration. Sensitivity of the Myrothecium particles to malonate, the most nearly specific inhibitor available, was typical of succinoxidase systems. Inhibition averaged about 50% at 0.0007 M giving a K_m/K_i value of about 60 which is similar to that of other highly active preparations under similar reaction conditions (21). Mycelial respiration was rather insensitive, averaging 0, 30, and 42% inhibition at 0.10, 0.23, and 0.39 M, respectively. This may indicate a permeability barrier although lowering the pH to 4 did not increase inhibition (3).

Because of the earlier work of Darby and Goddard (5, 6) on this organism it was of special interest to compare the sensitivity of the oxidase and of respiration to cyanide. Although there was considerable variability among preparations, the oxidase activity was somewhat more sensitive than the respiration (Table VI). The greater sensitivity to cyanide of our mycelium than that of Darby and Goddard, who found little or no inhibition at 0.001 M, is difficult to explain. Although both cultures were from the same original isolate (USDA 1344.2) strain differences may have been involved since variations in culture characteristics are not uncommon in our experience with this species. Furthermore, younger mycelium was used in the present work (24 instead of 48 hour cultures). It may also be possible that use of the Krebs center well mixture in the earlier work led to underestimation of cyanide sensitivity.

The response of Myrothecium particles to the alkylnapthoquinone, SN5949, which blocks cytochrome c reduction also was similar to that of mammalian succinoxidase preparations (2, 18, 24). Inhibition was complete at 3×10^{-6} M. Respiration was almost as sensitive, showing 85% inhibition at 3×10^{-6} M and 95% inhibition in saturated solutions, $4\text{-}6 \times 10^{-6}$ M. However, at less than 2×10^{-6} M, partial recovery of respiratory inhibition was observed (Fig. 3). The succinoxidase activity when tested with varying amounts of particles showed evidence of the irreversible (or pseudoirreversible) type described by Reif and Potter (18) for this compound with rat liver suspensions. This dependence of inhibition on particle concentration is shown in Fig. 4.

DISCUSSION

The principal object of this study was to establish a valid assay method and to determine the properties of the succinoxidase system in M. verrucaria mycelium. The extreme toughness of the fungal cell walls necessitated rather harsh grinding conditions which appeared to result in some damage to the isolated particles and in loss of cofactors. This was evidenced by pronounced variability in oxidative capacities of different particle preparations. Ability of particles to oxidize Krebs cycle intermediates including succinate was highly sensitive to washing, and even with addition of soluble cofactors marked variation in ability to oxidize pyruvate, ketoglutarate, fumarate, citrate, and malate was observed. Succinoxidase activity of unwashed particle preparations also varied considerably in response to the addition of cytochrome c and of ATP or glutamate and to variation in succinate concentration. Despite this variability, kinetic analysis indicated that use of ATP or glutamate permitted satisfactory estimates of maximum succinoxidase activity by direct measurement (ca. 95% of V_{max}).

The succinoxidase of the Myrothecium mycelial particles isolated as described was highly active and similar in general properties to those from other sources. The average QO_2 (N) value of 580 is above the 350 reported for Neurospora crassa extracts (20) and is comparable to the upper range of values for higher plant mitochondria (4, 11, 17). Such comparisons are difficult because of differences in reaction conditions and uncertainty as to how nearly optimum the assay conditions were, especially with respect to oxalacetate inhibition. It is also not always clear how much subsequent oxidations of the Krebs cycle may have affected the reported values for succinoxidase activity. As with other similar preparations, the mycelial oxidase was sedimented completely after 30 minutes at 20,000 x g; it was stimulated by added cytochrome c; and it was inhibited by dicarboxylic acids, cyanide, Cu, Co, and the alkylnaphthoquinone, SN5949. Stimulation of succinate oxidation by added ATP (11, 15, 16, 17) or glutamate (7) also is not unusual.

The increased activity of the fungal succinoxidase in the presence of ATP or glutamate could be attributed to removal of the competitive inhibitor, oxalacetate. The latter apparently was formed in concatenary reactions by malate oxidation at too slow a rate to be detected manometrically. The effect of glutamate can be explained by its role in the transamination of the oxalacetate to aspartate. The action of ATP, however, is more difficult to interpret.

The response to various adenine and uridine nucleotides suggested that "high-energy" phosphate bonds were involved in the stimulation of the succinoxidase system. This would require that AMP be phosphorylated by those particle preparations with which it was active. Attempts to demonstrate such phosphorylation by inorganic phosphate uptake were inconclusive. However, dinitrophenol at concentrations above 5×10^{-5} M partially or completely prevented AMP stimulation with several particle preparations which responded to the latter.

The effect of EDTA on adenylate stimulation indicated that metallic cations were involved. Though the identity of these remains uncertain, the small but consistent stimulation by Mg suggests that it may be the

essential ion. This cation has been reported to be effective in stimulating plant succinoxidase systems (11, 15, 17) and in protecting against added oxalacetate (1, 22).

Among possible mechanisms of ATP action, the further metabolism of oxalacetate, perhaps to phosphoenolpyruvate, was considered. An ATP-dependent CO_2 evolution with added oxalacetate was observed with one mycelial particle preparation, but this could not be repeated. In Tyler's detailed study (22) of the mechanism of ATP protection of rat tissue succinoxidase, no further metabolism of oxalacetate was observed. He proposed that an ATP-dependent metal-enzyme complex with oxalacetate was formed or that ATP and Mg might alter the particle so as to change the affinity for succinate and oxalacetate. The first proposal would appear to be more attractive than the second for the Myrothecium particle data. Some inhibition of the rat oxidase resulted from ATP addition, whereas only stimulation occurred with fungal particles. Furthermore, with the latter, ATP would reverse inhibition by fumarate, malate, oxalacetate, and Co, but it had no effect on inhibition by malonate or Cu. It would seem that a rather complex hypothesis would be required to explain such results on the basis of a physical change in particle structure. Another possible explanation of ATP stimulation might be based on the competitive inhibition of DPN function in the malic dehydrogenase system reported by Williams (25). This action of ATP also would reduce oxalacetate formation, but it would not explain ATP protection of the system with added oxalacetate.

The problem of relating in vitro succinoxidase activity to cellular function is a complicated one especially with fungus mycelium where extraction is difficult, permeability to inhibitors is uncertain, and indications of inhibitor detoxification are present. On the whole, however, the inhibitor evidence would suggest that the succinoxidase system has the same essential role in aerobic respiration in M. verrucaria that has been established in many other organisms. More conclusive evidence will require a more thorough study of related metabolic systems. While it seems likely that a Krebs cycle is present in M. verrucaria, further investigation of the metabolic properties of both particles and intact mycelium will be necessary to establish the fact.

SUMMARY

Particles from mycelium of Myrothecium verrucaria sedimented at 20,000 x g oxidized pyruvate, citrate, ketoglutarate, fumarate and malonate when fortified with cofactors. The succinoxidase system was stimulated by cytochrome c, glutamate, ADP, ATP, UTP, and in some preparations by AMP. A cation, prob bly Mg, was required for stimulation by nucleotides. The oxidase was inhibited by adding fumarate, malate, oxalacetate, Co, Cu, cyanide, malonate, and the alkylnaphthoquinone, SN5949. Inhibition by the latter appeared to be irreversible. ATP reversed inhibition by Co and the Krebs cycle acids. Stimulation by ATP and glutamate could be attributed to removel of oxalacetate formed by concatenary reactions. Under optimum conditions with glutamate and cytochrome c added, the K_m for succinate oxidation was 0.007 M and the average Q_{O_2} (N) was 580. The comparative sensitivity of mycelial

respiration and particle succinoxidase activity to malonate, cyanide, and SN5959 indicated an essential role for the enzyme system in respiration.

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THE EFFECTS OF THE EUROPEAN CORN BORER, PYRAUSTA NUBILALIS (HBN.), ON CORN YIELD[1]

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INTRODUCTION

In 1953 experiments were begun in Iowa, Minnesota and Ohio on a project entitled "Factors Influencing European Corn Borer Populations, Regional Project NC-20." The unique feature of these experiments was the use of the same experimental design and methods in all three states.

One problem posed by the experiments concerns the differentiation of the effects of two European corn borer broods in the three states. Present survey methods for estimating yield losses depend on fall surveys of borer abundance. Midsummer estimates of borer populations have been suggested for obtaining more accurate estimates of yield losses. The data considered here can perhaps shed some light on the results of estimating losses at various times in the season.

A second problem associated with the NC-20 data concerns the substitution of cavity counts for borer counts in the evaluation of damage. From an economic standpoint cavity counts in some situations would provide more information than borer counts.

Finally, there is interest in the assistance these data can provide for relating corn borer damage to loss in yield. Although the NC-20 experiments were established primarily for a study of ecological relations, the yield data collected are suitable for the estimation of yield losses.

REVIEW OF LITERATURE

In the United States some of the earliest work on losses in yield is reported by Caffrey and Worthley (1927). The average damage for the years 1920 to 1922 on experimental plots in the New England area showed 10.0, 3.14, and 1.4 per cent grain injury or destruction on flint, dent

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and sweet corn respectively. Up to 1924 there was little economic loss in Ohio and Michigan, the damage being confined mainly to New England. Caffrey and Worthley point out as early as 1928, in a supplement to their report, that studies demonstrate that indirect injury to cornstalks affecting number, weight and quality of ears are more important than the direct injury by larvae feeding on the ears.

Salter and Thatcher (1927) presented data bearing on losses in yield in Ohio for 1924 to 1926, noting that ears from infested stalks were 5.19 to 9.14 per cent lighter in weight than ears from noninfested stalks.

Beginning mainly with the work of Patch considerable use was made of regression methods to estimate loss per borer, either as a percentage or in bushels per acre per borer per plant. Artifical population levels were induced by applying varying numbers of egg masses to the plants.

Patch et al. (1938) in a comparison of resistance of two hybrids to a single generation borer attack reported a reduction in yield of 1.81 bushels per acre per borer per plant using Michigan 561, a resistant hybrid. Based on plantings from 1929 to 1932 a locally adapted Clarage variety, considered susceptible, had a yield reduction of 1.83 bushels per acre per borer per plant.

In a comparison of open pollinated varieties and hybrid field corn from 1930 to 1933 Patch et al. (1941) studied the effect of a single generation infestation on quality and yield of ears. Borer counts were made in mid-August while yield data were collected in October. Under infestations of up to five borers per plant there was little reduction in yield. Based on the estimated marketable yield in the absence of borers, the marketable yield was reduced 2.53 bushels (2.99 per cent) per borer per plant on hybrids, 2.45 bushels (3.71 per cent) on open-pollinated varieties, and 1.87 bushels (3.56 per cent) on the variety Clarage. The respective average estimated marketable yields in the absence of borers were 84.6, 66.1 and 52.4 bushels per acre.

Patch et al. (1942) studied the factors determining the reduction in yield using up to five infestation levels on field corn. Within the yield range of 28 to 85 bushels per acre the per cent loss in yield increased with increases in the normal yield. On Clarage, yield reduction was 1.37 bushels (4.86 per cent) per acre per borer per plant when yield in the absence of borers was 28.2 bushels, and 2.27 (2.68 per cent) when the yield was 84.6 bushels per acre. On hybrids the reduction was 2.57 bushels (3.02 per cent) per acre per borer per plant for a yield of 85 bushels and 4.13 bushels (3.93 per cent) per acre per borer per plant for a yield of 105 bushels per acre. The stage of plant development was found to be an important factor in yield reduction.

Studies of the loss of yield in dent corn hybrids infested with the August brood at Lafayette, Indiana, were reported by Deay et al. (1949) using 16 single crosses in 1944. The reduction of yield in bushels per acre at 15.5 per cent moisture for each additional borer per plant varied from .50 $\pm$ .221 to 2.46 $\pm$ .254 with an average value of 1.18 $\pm$ .069, or an estimated 1.85 per cent per borer per plant. An estimate of the yield in the absence of borers was computed for each single cross and it was found that the reduction in yield per borer was directly related to the yield of the hybrid, higher yielding varieties having a greater loss in yield. Additional experiments in 1945, using single crosses with P8

as a common parent, showed a loss in yield of 1.27 bushels per acre per borer per plant, whereas the WF9 crosses had a loss of 1.62 bushels per acre per borer per plant. Another group of P8 hybrids had a loss of 1.11 bushels while Hy and L317 groups lost 2.25 and 3.09 bushels per borer per plant respectively.

Chiang and Hodson (1950) point out four possible ways in which borer damage can occur: (1) interference with the plant physiology resulting from boring and feeding and consequent reduction in ear size, (2) destruction of kernels, (3) stalk breakage and ear droppage, and (4) provide entrance points for stalk rots. Their data suggested that first brood larvae caused stalk breakage above the ear, while ear dropping and breakage below the ear were caused mainly by the second brood.

Patch (1951) estimated the reduction in yield by the first brood, using four fields in Iowa, as 2.4 bushels (2.2 per cent) per borer per plant where the yield in the absence of borers was 108.8 bushels per acre. In this report, counts of cavities in the plant were utilized in estimating borer populations.

Stalk breakage by the August brood in Indiana in 1944 to 16 single cross hybrids was studied by Patch et al. (1951). The loss in yield due to reduction in ear size was estimated to be ten times the losses due to unrecoverable dropped ears and ears on broken stalks. Total reduction in yield was estimated as 1.87 per cent per borer per plant.

During 1952 Chiang, Cutkomp and Hodson (1954) found that combination of a low first brood borer population and a sizeable second brood did not significantly affect yield. Use of a 3 per cent loss per borer per plant, disregarding first and second brood borer effects, gave an overestimate of yield loss. It was suggested that use of first brood populations only would have given a better estimate. Second brood borers were found to have little effect on ear growth but were responsible for stalk breakage and ear dropping. It was concluded that the population of first and second brood borers must be distinguished and that their effect on the loss in yield should be assessed separately.

Using both cavity and borer counts, varying planting dates and varying artificial infestation levels Beck (1954) estimated damage in Iowa to a commercially grown susceptible hybrid, Iowa 4297. Cavities proved to be a better index of damage in the fall than borers. In a midseason planting, loss in yield based on first brood populations was 5.4 per cent per borer per plant and 2.6 per cent per cavity. In the fall, yield and borers had low correlation but yield and cavities were correlated. The loss in yield was 3.4 per cent per cavity in the fall. For populations made up of both broods loss per borer was 2.0 per cent and loss per cavity 1.99 per cent in the fall. On the second brood only, in a late planting of the same hybrid the loss per borer was 0.8 per cent and the loss per cavity was 0.9 per cent. There were indications that later applications of eggs resulted in less damage per borer per plant.

Of the papers reviewed up to this point, only two (Patch, 1950 and Beck, 1954) discuss the use of cavities in estimating yield losses. Both are unpublished and Patch only used cavities to estimate midsummer borer populations. Mention is made that the time of fall dissections affects the surviving borer population and consequent yield loss estimates per borer, particularly in two-generation areas.

EXPERIMENTAL DESIGN AND DATA

The data utilized here are from split-split plot experiments conducted from 1953 to 1956 in Iowa, Minnesota, and Ohio. A single experiment was conducted in each state each year. Main plot treatments consisted of planting dates, designated either early or late. The sub-plot treatments were a susceptible single cross (WF9 x M14), and a resistant single cross hybrid (Oh43 x Oh51A). The sub-sub-plot treatments, consisting of 42 hills of corn, were made up of eight combinations of first and second brood population levels. The designated treatments in the following table where N is a natural infestation, 3 or 6 refers to the addition of 3 or 6 egg masses manually applied in synchrony with the natural infestation. Control by either EPN (first brood) or DDT (second brood) is indicated by zero population.

		First brood level			
		0	N	N + 3	N + 6
		(Treatments)			
	0	1			
Second	N	5	2, 8	3	4
brood	N + 3	6			
level	N + 6	7			

At midseason six plants per plot were dissected and counts made of the larvae and cavities on treatments 1, 2, 3, and 4. A cavity is defined as a burrow in the stalk large enough to contain a full grown borer, or about 3/4 inch or more in extent. In the fall ten plants from every plot in the experiment were dissected to obtain borer and cavity counts, while in addition, total yield from 30 plants on each plot was obtained. The ten plants dissected for borer and cavity counts were included in the 30 plants on which yield data was obtained. The experiments were all designed and performed with six replicates. In three instances, however, only five replicates yielded usable data, namely in Iowa in 1954 and 1955 and in Minnesota in 1955.

The data were transferred to IBM punch cards after collection. The larvae and cavity totals on individual plants were then transformed by the square root transformation, a standard transformation for entomological counts. The value of $\sqrt{x + 0.5}$ where x is the number of borers or cavities per plant thus became the variable utilized in computations.

STATISTICAL STUDIES

Midsummer Dissections

Data from the midsummer dissections and the yield in the fall were used to estimate the regressions of yield on borers and yield on cavities. Four treatments, 1, 2, 3, and 4, were dissected at midsummer, but data from treatment 1 were omitted from the calculations since treatment 1 received insecticide applications during the second brood while the other three treatments did not.

Table 1. Correlation coefficients of yield and borers or cavities for various year, state, hybrid, date combinations (midsummer first brood dissections).

		ES[1]	ER	LS	LR
			Yield and borers		
Iowa	1953	-.663*	-.050	-.258	-.446
	1954	-.799*	-.474	-.738*	-.597*
	1955	-.570*	.116	-.554*	-.225
	1956	-.337	.015	-.806*	-.285
Minnesota	1953	-.002	.388	-.347	-.397
	1954	-.701*	.104	-.469	-.147
	1955	-.542**	-.407	-.891*	-.454
	1956	-.660*	-.541**	-.723*	-.311
Ohio	1953	-.414	.123	.450	.205
	1954	-.295	-.060	.182	.247
	1955	-.729*	-.490**	-.449	-.329
	1956	.091	.150	.138	-.259
			Yield and cavities		
Iowa	1953	-.683*	.025	-.341	-.352
	1954	-.840*	-.654*	-.786*	-.516**
	1955	-.759*	-.579*	-.434	.112
	1956	-.370	.082	-.832*	-.292
Minnesota	1953	-.152	.111	-.312	-.098
	1954	-.772*	-.161	-.249	-.317
	1955	-.566*	-.360	-.791*	-.596*
	1956	-.720*	-.496**	-.771*	-.290
Ohio	1953	-.376	.060	.359	.057
	1954	-.374	.128	.217	.490**
	1955	-.771*	-.407	-.312	-.317
	1956	.293	.208	.104	-.329

[1]E = early planting
L = late planting
S = susceptible variety
R = resistant variety

* indicates significance at the 5 per cent level.
** indicates significance at the 10 per cent level.

Each set of 15 or 18 observations for the various year, state, date of planting and variety combinations may be viewed as derived from a randomized block experiment with three treatments and either five or six replicates. If an analysis of covariance table using yield and borers or cavities is computed an estimate of the regression coefficient or correlation coefficient is obtained from the treatment plus error sums of squares. The success of estimating loss in yield knowing either the number of borers or cavities may be quickly seen from the correlation coefficients given in Table 1.

The results shown in Table 1 support the hypothesis of a relation between yield and some measure of infestation, either borer or cavity counts. In all cases of a statistically significant coefficient, except one, the coefficient is negative, indicating that higher infestations are associated with reduced yield. There is considerable variation from year to year and state to state in the number of instances when coefficients are significant. There is no evidence from the table values that borers are more closely associated with yield than cavities, either in terms of the number of significant coefficients or relative coefficient sizes. The most striking fact about the values in Table 1 is the apparently better chance of demonstrating a relation between yield and borers or cavities upon a susceptible variety (S) than upon a resistant variety (R). There is no indication that planting date has influenced the size of the correlations.

The use of the correlations is satisfactory for a rapid survey of the possibilities of demonstrating a relation between yield and some measure of population (borers or cavities). Of greater interest, however, is the estimation of regression equations.

A regression equation was computed for all cases of a significant negative correlation coefficient in Table 1. These equations are shown in Table 2.

There are no striking consistent differences in the regression coefficients which vary from -5.46 to -26.40. The yield value in these equations is in bushels per acre while the number of borers or cavities (x) is the square root of the actual number of borers or cavities per plant plus one-half. The value of x for one borer or cavity is therefore the square root of 1.50 or 1.22. For the yield and borers in Iowa, ES, 1953, the loss per one borer is therefore 8.23 bushels per acre while with ten borers the loss per borer would be 4.08 bushels. These results derive from the fact that the square root transformation was employed resulting in a curvilinear relation between yield and infestation. It should be remembered when computing the yield for a zero population of borers per plant that the value of x is not zero but 0.71 the square root of 0.50. When losses per borer are computed this adjustment must be made for the use of a $\sqrt{x + 0.5}$ transformation.

Treatment 1, as was stated at the beginning of this section, was not used for computing the estimating equations of Table 2 and it provides an independent estimate of the yield in the absence of borers. The mean yield on all plots with treatment 1 was 96.2 bushels per acre when averaged over all the factor combinations of Table 2 while the estimated mean yield using the estimating equations for zero borers (x = .71) was 97.5 bushels per acre. The mean actual loss (mean of treatment 1 minus mean of treatments 2, 3 and 4) was 9.86 bushels per acre while the mean

Table 2. Regression equations for midsummer first brood dissections relating yield in bushels per acre to borers or cavities per plant.

Factor combination			Yield and borers: Equation	Yield and borers: Standard error of b[1]	Yield and borers: d.f.[2]	Yield and cavities: Equation	Yield and cavities: Standard error of b	Yield and cavities: d.f.
ES	Iowa	1953	y = 110.52 - 16.14x	5.49	11	y = 105.52 - 7.56x	2.44	11
		1954	y = 120.91 - 16.98x	4.26	9	y = 125.69 - 16.86x	3.62	9
		1955	y = 105.07 - 18.36x	8.80	9	y = 114.67 - 18.36x	5.26	9
	Minnesota	1954	y = 116.04 - 12.42x	6.80	11	y = 125.81 - 26.40x	6.55	11
		1955	y = 100.31 - 11.64x	6.01	9	y = 96.47 - 5.46x	2.65	9
		1956	y = 111.96 - 8.76x	3.01	11	y = 115.20 - 9.96x	2.89	11
	Ohio	1955	y = 110.82 - 16.26x	4.61	11	y = 105.12 - 7.80x	1.94	11
ER	Iowa	1954				y = 103.45 - 17.52x	6.00	9
		1955				y = 114.26 - 15.54x	8.24	9
	Minnesota	1956	y = 109.91 - 14.10x	6.62	11	y = 108.82 - 12.12x	6.40	11
	Ohio	1955	y = 101.85 - 10.02x	5.37	11			
LS	Iowa	1954	y = 117.83 - 23.28x	7.10	9	y = 117.19 - 21.84x	5.73	9
		1955	y = 102.50 - 13.62x	6.85	9			
		1956	y = 111.23 - 15.30x	3.40	11	y = 115.12 - 19.68x	3.97	11
	Minnesota	1955	y = 88.25 - 21.24x	3.61	9	y = 81.70 - 12.66x	3.26	9
		1956	y = 104.97 - 10.38x	2.99	11	y = 106.94 - 10.86x	2.70	11
LR	Iowa	1954	y = 106.45 - 10.80x	4.83	9	y = 104.57 - 8.10x	4.47	9
	Minnesota	1955				y = 65.34 - 12.54x	5.62	9

[1] b is the regression coefficient.

[2] d.f. = degrees of freedom.

estimated loss was 9.50 bushels per acre. With cavities, the yield on treatment 1 was 93.8 bushels per acre while the mean for treatments 2, 3, and 4 was 96.7 bushels per acre. The actual loss was 9.64 bushels per acre while the estimated loss was 11.55 per acre.

The correlation coefficients for actual and estimated yields and actual and estimated yield losses for the equations of Table 2 using borers are 0.845 and 0.703 respectively with 13 degrees of freedom, and 0.841 and 0.612 respectively with 14 degrees of freedom using cavities. These correlations are all significant at the 5 per cent level and indicate that the estimated yields or yield losses tend to vary as the actual yields or yield losses.

Having computed the equations shown in Table 2 as well as having tested the significance of the correlation coefficients shown in Table 1 the question which naturally arises is whether or not the data can be pooled in order to reduce the number of estimating equations. The preceding results are based on the best, in the sense of being significant at the 5 per cent level (in six cases 10 per cent level) set of equations obtainable from the original data. Larger regression coefficients or maximum loss estimates per borer or cavity in general will be the result. The remaining portions of this section consider the pooling of all the data from the various hybrid-planting date data combinations over years within Iowa and Minnesota. As a result of this utilization of all data the regression coefficients and loss estimates per borer will generally be lower.

There are a number of ways in which the pooling and particularly tests for the homogeneity of the regressions can be made. For the data considered here the question asked is whether for each planting date, variety combination in a state the regression coefficients are relatively constant from year to year. Ostle (1954) presents methods for performing various statistical tests to answer this question and consequences of it. These tests were applied to the four regressions for each planting date, hybrid, year combination.

There were no instances with the Ohio data where a pooled regression showed significance with either borers or cavities. Of the four coefficients for each variety-date combination some were positive and some negative. Generally the coefficients over the four years were homogeneous but in no case was the pooled coefficient significantly different from zero.

For the Minnesota data the same situation prevailed on the early planted-resistant hybrid combination. With the other three date-hybrid combinations the regression coefficients from year to year were considered homogeneous except with cavities on early-susceptible (ES) in 1954 when the regression coefficient was rather high (-26.40) as compared to the other three coefficients.

With the Iowa data for the early-susceptible combination the results were similar to those for Minnesota. The early-resistant combination using borers and yield gave no indication of a yield loss relation. The equations for the late-susceptible combination in Iowa were the most consistent from year to year of those equations for any date-hybrid combination in either Iowa or Minnesota with both borers and cavities with yield. The regression coefficients for the late-resistant equations were homogeneous.

The problem to be considered next concerns the estimation of yield loss regression coefficients for the planting date-hybrid combinations pooling the data obtained over four years although in some instances there was evidence that regressions were nonhomogeneous. These final results on the first brood populations are shown in Tables 3 and 4.

Comparisons of the ratio of the standard error relative to the respective regression coefficient in Table 3 are of assistance in assaying the efficiency of borers and cavities for determining yield losses. These ratios for the same hybrid-date combination are approximately the same and would indicate that cavities are suitable criteria for evaluating damage as compared to borer counts, in terms of relative variation.

Table 3 contains the estimating equations when pooled over years. In all cases the coefficients are significant at the 5 per cent level except the late-resistant combination in Minnesota which is significant at the 10 per cent level. The unweighted means of the coefficients are -10.22 for borers and yield and 9.78 for cavities and yield. The values result in losses of 5.21 bushels per acre at a mean of one borer per plant or 4.99 bushels per acre at a mean of one cavity per plant. As was anticipated, these values are lower than the respective estimates of 7.57 and 7.32 bushels loss using only regressions for Iowa and Minnesota which were significant.

Table 3. Yield loss equations in bushels per acre per borer per plant pooled over years for midsummer dissections.

Factors		Equation	Standard error of b	d.f.
		Yield and borers		
Iowa	ES	$y = 98.97 - 11.16x$	2.65	43
	LS	$y = 105.53 - 13.47x$	2.44	43
	LR	$y = 96.86 - 8.00x$	2.98	43
Minnesota	ES	$y = 116.75 - 8.62x$	2.17	45
	LS	$y = 108.61 - 10.70x$	2.81	45
	LR	$y = 91.28 - 9.35x$	5.11	45
		Yield and cavities		
Iowa	ES	$y = 101.98 - 10.78x$	2.23	43
	ER	$y = 95.20 - 7.93x$	2.90	43
	LS	$y = 105.99 - 13.25x$	2.41	43
	LR	$y = 94.57 - 5.27x$	2.83	43
Minnesota	ES	$y = 118.03 - 10.07x$	2.44	45
	LS	$y = 108.60 - 11.57x$	3.67	45
	LR	$y = 91.32 - 9.59x$	5.40	45

Table 4 demonstrates the effect of the square root transformation on the loss equations. As a consequence of the transformation, loss per borer or cavity depends upon the number of borers or cavities. The losses in bushels per acre per borer or cavity per plant when 1, 3, or 5 per plant are present in terms of the 95 per cent confidence intervals are shown. The data in this table point out the fact that depending on the population range, various loss estimates can be obtained for loss in yield per borer or cavity when a linear relation is assumed.

Finally, the equations shown in Table 3 were employed to determine the lines shown in Figs. 1, 2, 3, and 4. For Iowa there is not much difference in the lines in terms of slopes or distances between lines for the same borer population. In Minnesota the slopes are approximately the same but considerable differences in yield for the same borer population are present, depending on the planting date-hybrid combination. The situations demonstrated by these graphs indicate that for these data a single coefficient expressed as a percentage loss per borer per plant might be reasonable in Iowa. However, in Minnesota any percentage loss figure would depend on the estimated yield in the absence of borers since the regression coefficients are relatively constant. Considerable errors in loss estimates would obtain unless a scheme taking yield into account were employed.

Table 4. Ninety-five percent confidence limits of loss in bushels per acre per borer per plant or cavity under various infestations for midsummer dissections.

Infestation level		ES	LS	LR	ES	ER	LS	LR
		Yield and borers			Yield and cavities			
Iowa								
1 per plant	L_1	2.97	4.36	1.01	3.21	1.06	4.28	-0.21
	L_2	8.42	9.39	7.13	7.79	7.03	9.24	5.60
3 per plant	L_1	2.25	3.30	0.77	2.43	0.81	3.25	-0.16
	L_2	6.39	7.11	5.41	5.91	5.34	7.01	4.25
5 per plant	L_1	1.90	2.79	0.65	2.06	0.68	2.75	-1.37
	L_2	5.40	6.02	4.57	5.00	4.51	5.92	3.59
Minnesota								
1 per plant	L_1	2.17	2.57	-0.48	2.63		2.13	-0.66
	L_2	6.61	8.35	10.01	7.64		9.67	10.43
3 per plant	L_1	1.65	1.95	-0.37	2.00		1.63	-0.49
	L_2	5.02	6.34	7.59	5.79		7.34	7.92
5 per plant	L_1	1.39	1.65	-0.31	1.69		1.37	-0.41
	L_2	4.24	5.36	6.42	4.90		6.20	6.69

STATISTICAL STUDIES

Fall Dissections

In the fall, data from dissections of corn plant samples as well as yield estimates from every plot in each experiment were collected. The data for the eight treatments or levels of infestation within a planting date-variety combination can be conveniently separated into two groups for purposes of further analysis. Treatments 2, 3, 4, and 5 represent four levels of a first brood with a natural second brood population, while the combination of treatments 1, 5, 6, and 7 represents four levels of the second brood in the relative absence of a first brood.

The analyses to be considered in this section are a natural extension of those in the section on midsummer first brood dissections. Two broods instead of one are considered and four levels of infestation (treatments) instead of three are usable. The Ohio data for 1955 have been omitted from the succeeding analyses since borers or cavities for the fall dissections were not recorded on a per plant basis.

Tables 5, 6, and 7 present the correlation coefficients for Iowa, Minnesota, and Ohio. For years when five replicates were used the coefficients are based on 14 degrees of freedom and when six replicates were used the coefficients are based on 17 degrees of freedom. As in the case of midsummer dissection results, the treatment plus error sums of squares and cross-products were used in the computations.

These results from fall dissections should be compared with the results from the midsummer dissections of treatments 2, 3, and 4 shown in Table 1. Consideration of the Iowa data (Table 5) for first brood with Iowa results in Table 1 indicates that the time of dissection for the estimation of yield loss equations may have considerable effect on the chances of demonstrating a relation between yield and a measure of the infestation. With the midsummer dissection data there was no evidence that borers were superior to cavities for estimating yield losses in terms of significance of correlation coefficients. However, when dissections were performed in the fall after the natural second brood has had an effect, correlations of yield and borers were negative in only two instances out of sixteen. The positive correlations were even significant in five instances. This result indicates that if a research worker were trying to establish loss equations using yield and borers obtained from fall dissections, he would very likely be unsuccessful and also that a 3 per cent loss per borer based on fall dissection would have been in error in Iowa.

If cavities had been used in Iowa for establishing loss equations based on fall dissections, greater success might be anticipated. Using cavities and yield, 11 instances of a negative coefficient occur, and five are significant. When confronted with both midsummer and fall results, the midsummer dissections for estimating yield losses would be considered superior to fall dissections in terms of higher correlation coefficients.

The more frequent occurrence of significant positive correlation coefficients is a very disconcerting event with the fall dissection data. For midsummer data positive coefficients may be attributed to failures in establishing infestation levels and yield variability. Fall infestations are also subjected to the interaction of infestations of each brood.

Table 5. Correlation coefficients of yield and borers or cavities for various year, hybrid, date combinations in Iowa for fall dissections of both broods.

	ES	ER	LS	LR	ES	ER	LS	LR
	Yield and borers				Yield and cavities			
	Levels of first brood (treatments 2, 3, 4, 5)							
1953	.234	.227	.305	.261	-.193	-.155	-.305	.100
1954	.732*	.320	.848*	.240	-.679*	-.011	.708*	.056
1955	.477**	.589*	.496*	.178	-.590*	.462**	-.157	-.262
1956	-.094	.292	-.408**	.353	-.425**	.020	-.787*	-.362
	Levels of second brood (treatments 1, 5, 6, 7)							
1953	-.522*	-.298	-.242	-.671*	-.431**	-.244	-.239	-.635*
1954	-.536*	-.667*	-.764*	-.426**	-.593*	-.625*	-.727*	-.460**
1955	-.177	.098	-.436**	.009	-.243	.393	-.389	-.152
1956	-.302	-.525*	-.635*	-.784*	-.276	-.531*	-.643*	-.818*

Table 6. Correlation coefficients of yield and borers or cavities for various year, hybrid, date combinations in Minnesota for fall dissections of both broods.

	ES	ER	LS	LR	ES	ER	LS	LR
	Yield and borers				Yield and cavities			
	Levels of first brood (treatments 2, 3, 4, 5)							
1953	-.122	-.243	.034	.006	.050	.035	-.139	.167
1954	-.601*	-.096	-.531*	.133	-.781*	-.110	-.484*	-.082
1955	.359	-.038	.185	.363	-.660*	.026	-.314	-.136
1956	-.524*	-.136	-.568*	.064	-.632*	-.370	-.744*	.183
	Levels of second brood (treatments 1, 5, 6, 7)							
1953	-.173	-.323	-.121	-.303	.061	-.351	.004	-.120
1954	.312	-.070	.169	-.204	.376	-.089	.195	-.185
1955	.090	-.241	-.367	.046	.007	-.297	-.354	-.046
1956	-.389**	-.247	-.256	.524*	-.542*	-.131	-.419**	-.114

Table 7. Correlation coefficients of yield and borers or cavities for various year, hybrid, date combinations in Ohio for fall dissections of both broods.

	ES	ER	LS	LR	ES	ER	LS	LR
	Yield and borers				Yield and cavities			
	Levels of first brood (treatments 2, 3, 4, 5)							
1953	-.703*	-.219	-.046	.286	-.689*	-.101	.139	.309
1954	-.194	.135	.068	.213	-.134	-.049	-.211	.065
1956	-.580*	-.047	-.442**	-.314	-.523*	-.328	-.479*	-.352
	Levels of second brood (treatments 1, 5, 6, 7)							
1953	-.375	-.712*	-.759*	-.556*	-.377	-.725*	-.777*	-.513*
1954	-.596*	-.579*	-.832*	-.799*	-.614*	-.596*	-.836*	-.872*
1956	-.489*	-.676*	-.419**	-.628*	-.588*	-.635*	-.545*	-.537*

Positive correlations between yield and borers at the time of fall dissections might occur if an inverse relation existed between infestations for first brood and for second brood. Evidence for an inverse relation between the two broods is available (Everett et al. 1958; Weekman 1957) and would partially explain higher second brood borer populations on plants with lower first brood populations. Thus when high first brood populations cause reduced yield they also tend, in a sense, to reduce second brood populations. The result would be a positive correlation between yield and fall population borers.

The fact that cavities at the time of fall dissections are more often negatively correlated with yield than borers would suggest that the cavity per borer per plant output for the second brood is not as high as for the first brood. Maturity of the plant would be associated with the ease with which cavities are produced and consequently the cavities produced by the second brood would not be sufficient in number to eliminate the effects of first brood cavities when correlated with yield.

First brood studies in Minnesota are not as striking as in Iowa. Midsummer data (Table 1) tended to give higher, and more often negative correlations of borers or cavities with yield than fall data (Table 6) for the first generation. It is noteworthy that for late plantings which would tend to be more heavily infested by natural populations, all eight coefficients for either borers or cavities in Table 1 are negative. The coefficients in Table 6 on late planting for levels of first brood are positive six out of eight times using borers and yield and negative six out of eight times with cavities. Once again for evaluating loss in yield due to first brood infestation it would appear that midsummer dissections were superior to fall dissections.

First brood studies in Ohio by means of fall dissections appear as fruitless as by the means of midsummer dissections. Coefficients were low except on the early planting of the susceptible variety (Tables 1, 7).

The studies on second brood levels in Tables 5, 6, and 7 are based on treatments 1, 5, 6, and 7. For Minnesota (Table 6) the results resemble those in Ohio for first brood. Very few instances of significant correlations were found. There was no indication that cavities were more highly correlated with yield than were borers.

The second brood data for Iowa (Table 5) indicates that loss equations could be determined and that the second brood appears to extend an effect on yield. It will be seen later, however, that losses per borer or cavity are not as great as in the case of first brood populations. No indication of a higher correlation between yield and borers than between yield and cavities to be observed.

The second brood data for Ohio shown in Table 7 present an extreme in the possibility of estimating yield loss. Every coefficient is negative and in only one case, the early-planted susceptible in 1953, is the coefficient not significant. Both borers and cavities appear to be equally effective in accounting for variation in yield.

For each instance of a significant correlation coefficient in Tables 5, 6, and 7 the estimating loss equation has been computed and is shown in Tables 8, 9, and 10. Instances where the regression coefficient is positive as occurs with first brood levels (treatments 2, 3, 4, and 5) have been included. The loss equations in these instances only appear in Table 8 for Iowa and are of no value in estimating yield reductions. The regression coefficients for the second brood in Iowa tend generally to be lower than those of the first brood.

The equations with a positive value for the regression coefficient are of no value in estimating yield loss but fit the collected data over a short range of values. It should be recalled that a parabola is being fitted to the data and that only a short segment of the entire curve is being utilized. Extrapolation without regard to the range of values of x and to the fact that x is actually a square root produces rather absurd results. The few instances for Iowa when the equations appear reasonable occur when cavities and yield are used.

Generally the coefficients for fall dissections of second brood (treatments 1, 5, 6, 7) are smaller than those of Table 2 for the first brood unaffected by a natural second brood, indicating that on a per borer or per cavity basis the first brood causes larger reductions in yield than the second brood.

The regression equations for data from Minnesota are shown in Table 9. The fact that there was relatively little success in estimating loss resulting from second brood infestation combined with some degree of success in estimating losses from first brood at the fall dissection is in contrast to the results in Iowa when the reverse was true. The absence of significant regression relations on the resistant hybrid is also noteworthy.

The equations for the Ohio data on borers or cavities and yield for first brood infestations shown in Table 10 show the regression coefficients to be significant in only three cases out of a possible 12. The results are approximately the same as for midsummer dissections. Considerable success was achieved in obtaining significant regression coefficients with second brood populations.

Table 8. Regression equations for fall dissections relating yield in bushels per acre to borers or cavities per plant in Iowa.

Treatments	Factors	Year	Equation	Standard error of b	d.f.	Equation	Standard error of b	d.f.
			Yield and borers			Yield and cavities		
2,3,4,5	ES	1954	y = 8.43 + 32.55x	8.10	14	y = 242.43 - 41.87x	12.10	14
		1955	y = 57.89 + 18.52x	9.11	14	y = 93.60 - 7.31x	2.67	14
		1956				y = 109.59 - 9.97x	5.14	17
	ER	1955	y = 53.51 + 23.80x	8.73	14	y = 62.55 + 11.37x	5.83	14
	LS	1954	y = - 6.35 + 39.55x	6.61	14	y = -53.08 + 35.25x	9.41	14
		1955	y = 70.95 + 12.28x	5.74	14			
		1956	y = 146.38 - 24.11x	13.10	17	y = 151.60 - 19.06x	3.62	17
1,5,6,7	ES	1953	y = 105.29 - 5.65x	2.24	17	y = 102.97 - 2.61x	1.33	17
		1954	y = 112.62 - 4.83x	2.03	14	y = 115.23 - 4.51x	1.64	14
	ER	1954	y = 104.09 - 3.84x	1.14	14	y = 104.23 - 3.33x	1.11	14
		1956	y = 100.10 - 3.02x	1.18	17	y = 99.53 - 2.36x	0.91	17
	LS	1954	y = 131.29 - 10.69x	2.41	14	y = 128.60 - 7.89x	1.99	14
		1955	y = 96.79 - 5.36x	2.96	14			
		1956	y = 121.88 - 6.63x	1.96	17	y = 121.39 - 5.28x	1.53	17
	LR	1953	y = 103.51 - 7.62x	2.04	17	y = 101.25 - 4.78x	1.41	14
		1954	y = 107.02 - 3.26x	1.85	14	y = 109.07 - 3.30x	1.70	14
		1956	y = 118.47 - 6.26x	1.20	17	y = 118.14 - 5.37x	0.92	17

Table 9. Regression equations for fall dissections relating yield in bushels per acre to borers or cavities per plant in Minnesota.

Treatments	Factors	Year	Equation	Standard error of b	d.f.	Equation	Standard error of b	d.f.
			Yield and borers			Yield and cavities		
2,3,4,5	ES	1954	y = 141.73 - 43.45x	12.95	17	y = 121.03 - 12.46x	2.41	17
		1955				y = 120.87 - 17.09x	5.19	14
		1956	y = 128.67 - 30.09x	11.86	17	y = 118.02 - 10.62x	3.16	17
	LS	1954	y = 123.40 - 33.16x	12.82	17	y = 104.03 - 8.02x	3.52	17
		1956	y = 115.02 - 25.17x	8.84	17	y = 111.32 - 11.55x	2.52	17
1,5,6,7	ES	1956	y = 116.75 - 13.75x	7.90	17	y = 113.59 - 6.70x	2.52	17
	LR	1956	y = 61.11 + 29.16x	11.48	17			
	LS	1956				y = 103.56 - 4.99x	2.62	17

Table 10. Regression equations for fall dissections relating yield in bushels per acre to borers or cavities per plant in Ohio.

Treatments	Factors	Year	Equation	Standard error of b	d.f.	Equation	Standard error of b	d.f.
			Yield and borers			Yield and cavities		
2,3,4,5	ES	1953	$y = 120.04 - 37.76x$	9.27	17	$y = 102.72 - 17.38x$	4.43	17
		1956	$y = 139.19 - 24.40x$	8.31	17	$y = 128.47 - 8.80x$	3.48	17
	LS	1956	$y = 110.65 - 12.92x$	6.35	17	$y = 108.26 - 6.62x$	2.95	17
1,5,6,7	ES	1954	$y = 93.57 - 8.53x$	2.79	17	$y = 93.41 - 7.47x$	2.33	17
		1956	$y = 139.51 - 31.84x$	13.77	17	$y = 132.45 - 17.04x$	5.68	17
	ER	1953	$y = 92.36 - 12.17x$	2.91	17	$y = 92.57 - 10.69x$	2.47	17
		1954	$y = 96.57 - 10.21x$	3.48	17	$y = 95.44 - 8.62x$	2.82	17
		1956	$y = 130.31 - 23.08x$	6.11	17	$y = 122.50 - 11.95x$	3.52	17
	LS	1953	$y = 86.57 - 7.41x$	1.54	17	$y = 86.84 - 6.81x$	1.34	17
		1954	$y = 110.21 - 16.33x$	2.64	17	$y = 108.57 - 13.46x$	2.14	17
		1956	$y = 116.05 - 16.44x$	8.63	17	$y = 117.66 - 13.95x$	5.21	17
	LR	1953	$y = 80.17 - 3.72x$	1.35	17	$y = 79.63 - 3.09x$	1.26	17
		1954	$y = 101.78 - 13.14x$	2.40	17	$y = 102.23 - 12.05x$	1.64	17
		1956	$y = 120.50 - 26.37x$	7.93	17	$y = 109.05 - 11.05x$	4.20	17

The results given in Tables 2, 8, 9, and 10 strongly support biological evidence that yield loss situations are different in the three states. This suggests that some modifications of the sampling time would be necessary to improve the estimates of yield loss. In situations where one brood generally overshadows the effect of the other brood, as at the locations in Ohio and Minnesota where these experiments were conducted, a fall sample of either borers or cavities (cavities preferred when the first brood is of greater significance) would be reasonable. Where the combination and interactions of broods occurs a midsummer first brood dissection counting either borers or cavities is indicated. In the absence of a midsummer dissection more importance should be placed on fall cavity counts.

The computation of loss equations pooled over years was considered next. For first brood borer levels there was no indication that a pooled regression on the resistant hubrid would be statistically significant in any of the states. On the susceptible hybrid in Ohio and Minnesota the regression coefficients from year to year were homogeneous and pooled equations were computed. For Iowa the only pooled regression equation was computed for the early-susceptible combination using cavities.

The regression coefficients from year to year with the second brood infestation levels were generally accepted as being homogeneous in Iowa and Minnesota. In Ohio there was evidence of heterogeneity which is believed to reflect the effects of year to year fluctuations in borer survival. The pooled regression equations based on the fall dissection data are shown in Tables 11 and 12.

The pooled equations for first brood studies (treatments 2, 3, 4, 5) are shown in Table 11. The equations in this table may be compared with those shown in Table 3. The success in obtaining pooled equations on the susceptible hybrid is in contrast to the lack of success when using the resistant hybrid. The fact that it was not possible to obtain a significant pooled regression using yield and borers from the first brood levels with fall dissections in Iowa is important for current loss estimation methods depend on a fall dissection.

The estimation of yield loss depends on the assumption that the rate of loss coefficient in an area being surveyed is the same as the rate of loss coefficient determined under experimental conditions. The rate of loss coefficient is in turn related to the time of dissection. An assumption of the same loss rates implies that dissections are performed at the same relative maturity of the borer population. A rate of loss coefficient determined from dissections in the early fall would be expected to be smaller than one determined in the late fall, particularly if the two coefficients were based on borer counts. The diminishing borer population in the fall would account for this difference in coefficients. Cavities, however, would be a more stable criterion in that they would not be subject to reduction in number as the season progressed.

The lack of significance for the Iowa regressions averaged over the four year period based on fall dissections of first brood infestations presents no evidence of a consistent regression of yield on borers or cavities. The application of a 3 per cent loss per borer value would not properly estimate the true yield loss. In order to better estimate loss in yield a dissection time during which the regression relation holds should be determined. Clearly the midsummer first brood dissection results would

Table 11. Yield loss equations in bushels per acre pooled over years for first brood levels (treatments 2, 3, 4, 5) based on fall dissections.

State	Factors		Equation	Standard error of b	d.f.
			Yield and borers		
Iowa	ES	p[1]			
	ER	p			
	LS	p			
	LR	p			
Minnesota	ES		$y = 124.51 - 23.24x$	8.47	68
	ER	ns			
	LS		$y = 108.34 - 17.44x$	7.23	68
	LR	ns			
Ohio	ES		$y = 115.91 - 23.56x$	5.07	53
	ER	ns			
	LS		$y = 100.34 - 10.24x$	4.48	53
	LR	ns			
			Yield and cavities		
Iowa	ES		$y = 108.38 - 10.04x$	2.75	65
	ER	p			
	LS	h			
	LR	ns			
Minnesota	ES		$y = 119.49 - 11.01x$	2.04	68
	ER	ns			
	LS		$y = 105.26 - 9.16x$	2.36	68
	LR	ns			
Ohio	ES		$y = 104.92 - 9.39x$	2.25	53
	ER	ns			
	LS		$y = 97.43 - 6.06x$	2.24	53
	LR	ns			

[1]These equations were omitted for the following reasons:

p - The pooled regression coefficient was positive.
ns - The pooled regression coefficient was non-significant at the 5 percent level.
h - The within year regression coefficients were non-homogeneous.

Table 12. Yield loss equations in bushels per acre pooled over years for second brood levels (treatments 1, 5, 6, 7) based on fall dissections.

State	Factors		Equation	Standard error of b	d.f.
			Yield and borers		
Iowa	ES		y = 96.20 - 3.74x	1.18	65
	ER		y = 90.29 - 2.56x	0.73	65
	LS		y = 106.89 - 6.82x	1.23	65
	LR		y = 98.62 - 3.97x	0.87	65
Minnesota	ES	ns			
	ER		y = 101.70 - 10.22x	4.69	68
	LS	ns			
	LR	ns			
Ohio	ES		y = 99.97 - 8.12x	2.56	53
	ER		y = 104.84 - 13.19x	2.26	53
	LS		y = 101.62 - 10.64x	1.71	53
	LR		y = 94.70 - 7.59x	1.63	53
			Yield and cavities		
Iowa	ES		y = 95.66 - 2.47x	0.82	65
	ER		y = 89.59 - 1.77x	0.56	65
	LS		y = 104.81 - 4.32x	0.88	65
	LR		y = 99.00 - 3.34x	0.66	65
Minnesota	ES	ns			
	ER		y = 99.92 - 6.90x	3.74	68
	LS	ns			
	LR	ns			
Ohio	ES		y = 101.53 - 7.67x	1.90	53
	ER		y = 103.46 - 10.37x	1.69	53
	LS		y = 102.54 - 9.69x	1.38	53
	LR		y = 95.13 - 6.83x	1.32	53

have satisfied the need for a significant yield with borers or cavities relation.

The pooled equations for the second brood dissections in the fall are shown in Table 12. All pooled equations in Iowa whether on cavities or borers were significant at the 5 per cent level. The regression coefficients are lower than those for the first brood regression coefficients determined from midsummer dissections.

Only two pooled equations were computed for the Minnesota data (Table 12) and these were on the early-planted resistant combination for yield and both borers and cavities. In general it would appear that second brood effects on yield in Minnesota are less pronounced than in Ohio or Iowa.

As in the case of the midsummer first brood dissection data, the 95 per cent confidence limits of loss per borer have been computed for the fall dissections. These limits are shown in Table 13 for the levels of the first brood and in Table 14 for the second brood levels.

The 3 per cent loss per borer figure based on Ohio data corresponds to losses per acre of 2.10 bushels per borer per plant when the yield is 70 bushels and 3.30 bushels per borer per plant when the yield is 110 bushels per acre. With the Ohio data in Table 14 for populations of three borers per plant these values of 2.10 and 3.30 are seen to lie within the confidence intervals, which are rather wide. However, for Iowa the values appear to be somewhat high, indicating overestimates of loss when only the second brood is involved. When only a first brood population is involved with midsummer dissections as an index as shown in Table 4 the established values based on the 3 per cent value appear to be too low for the susceptible hybrid in Iowa.

Table 13. Ninety-five percent confidence limits of loss in bushels per acre per borer or cavity per plant with various infestation levels for fall dissection of first brood levels (treatments 2, 3, 4, 5).

State	Infestation level		Yield and borers ES	Yield and borers LS	Yield and cavities ES	Yield and cavities LS
Iowa	1 per plant	L_1			2.37	
		L_2			8.08	
	3 per plant	L_1			1.76	
		L_2			6.01	
	5 per plant	L_1			1.48	
		L_2			5.07	
Minnesota	1 per plant	L_1	3.30	1.56	3.61	2.31
		L_2	20.87	16.58	7.84	7.21
	3 per plant	L_1	2.45	1.16	2.69	1.72
		L_2	15.53	12.34	5.84	5.37
	5 per plant	L_1	2.07	0.98	2.27	1.46
		L_2	13.13	10.42	4.93	4.53
Ohio	1 per plant	L_1	6.97	0.65	2.54	0.82
		L_2	17.54	10.00	7.23	5.49
	3 per plant	L_1	5.19	0.48	1.89	0.61
		L_2	13.06	7.44	5.38	4.08
	5 per plant	L_1	4.38	0.41	1.60	0.51
		L_2	11.03	6.29	4.54	3.45

Table 14. Ninety-five percent confidence limits of loss in bushels per acre per borer or cavity per plant with various infestation levels for fall dissection of second brood levels (treatments 1, 5, 6, 7).

State	Infestation level		ES	ER	LS	LR	ES	ER	LS	LR
			Yield and borers				Yield and cavities			
Iowa	1 per plant	L_1	0.72	0.57	2.27	1.16	0.43	0.34	1.33	1.05
		L_2	3.17	2.09	4.83	2.97	2.14	1.50	3.16	2.42
	3 per plant	L_1	0.53	0.43	1.69	0.86	0.32	0.25	0.99	0.78
		L_2	2.36	1.56	3.59	2.21	1.59	1.12	2.35	1.80
	5 per plant	L_1	0.45	0.36	1.43	0.73	0.27	0.21	0.84	0.66
		L_2	1.99	1.31	3.03	1.87	1.34	0.95	1.99	1.52
Minnesota	1 per plant	L_1		0.45				-0.29		
		L_2		10.18				7.47		
	3 per plant	L_1		0.33				-0.22		
		L_2		7.58				5.56		
	5 per plant	L_1		0.28				-0.18		
		L_2		6.40				4.70		
Ohio	1 per plant	L_1	1.55	4.50	3.75	2.25	2.01	3.63	3.60	2.17
		L_2	6.90	9.21	7.32	5.65	5.97	7.16	6.48	4.93
	3 per plant	L_1	1.15	3.35	2.79	1.67	1.49	2.70	2.68	1.62
		L_2	5.13	6.86	5.45	4.20	4.44	5.33	4.82	3.67
	5 per plant	L_1	0.97	2.83	2.36	1.41	1.26	2.28	2.26	1.37
		L_2	4.34	5.79	4.60	3.55	3.75	4.50	4.07	3.10

The pooled regression equations shown in Tables 12 and 13 have been plotted and are shown in Figs. 5 to 12. The equation for the Iowa data with yield and cavities in Table 11 (first brood) and the Minnesota equations in Table 12 (second brood) have not been included. These figures are directly comparable to Figs. 1 to 4 based on midsummer first brood dissections.

The figures for Minnesota are especially interesting since clear comparisons of a midsummer to a fall dissection of first brood infestations are possible. The figures based on the midsummer dissection (Figs. 3 and 4) show little difference between the use of borers or cavities. However, Figs. 5 and 6 show a large difference in the slope of the regression line for borers and yield as compared to cavities and yield. The difference between the slope of the lines in Fig. 5 as compared to the slope of the lines in the other three figures is thought to reflect the inconsistency of borer counts from midstream to fall and the consistency of cavity counts. The figures for borers show the effect of the reduction in the population in the fall upon the regression coefficients for as the population decreased the slope or estimated loss per borer would increase. The same size error in the estimation of the number of borers leads to a larger error in estimation of yield loss when using Fig. 5 than when using Fig. 3 as a result of the greater slope.

The equations for Ohio data are shown in Figs. 7 to 10. The second brood equations show little differences in slope associated with the hybrid-planting date combination. The first brood equations differ considerably when borers and yield are used. The Iowa results in Figs. 1, 2, 11, and 12 indicate differences in the rate of yield loss depending on the brood. There are no differences indicated in the effectiveness of borers or cavities for estimating loss in yield.

The final comparisons considered were of the relative variations for the estimates of the regression coefficients. The ratios of the standard error of the regression coefficient to the regression coefficient were computed for all situations in Tables 2, 8, 9, and 10 when an equation using yield and borers was paralleled by an equation using yield and cavities on the same combinations of year, planting data, variety and state. These ratios for borers were not consistently lower or higher than for cavities. This result leads to the conclusion that in terms of variability of the regression coefficients borers are neither better nor worse than cavities for estimating yield losses. Also, it may be observed that in almost every instance of a significant regression between yield and borers the parallel regression of yield and cavities is significant.

SUMMARY AND CONCLUSIONS

Data from split-split plot experiments with two hybrids, two planting dates and eight combinations of first and second brood infestations conducted at Ankeny, Iowa, Waseca, Minnesota, and Wooster, Ohio from 1953 to 1956 were considered for study by use of regression methods of the loss in yield of corn relative to the number of borers or cavities. Midsummer dissections were made to obtain an estimate of the first brood infestation and fall dissections were used in estimating the number of borers and cavities resulting from both broods.

The most important point established by the data was the inconsistency from state to state in the relative importance of the two broods to corn yield. In Ohio first brood effects were generally small while losses resulting from second brood attacks were pronounced. In Minnesota the opposite situation was revealed, major losses being caused by first brood populations. The data for Iowa indicated an intermediate situation with the interaction of the two broods affecting the estimation of losses.

The widely used 3 per cent loss figure was consistent with the loss estimates for Ohio and the Minnesota data. In Iowa for fall dissections estimates of a second brood the figure was too high, while for midsummer dissections estimates of the first brood on a susceptible hybrid the figure was too low.

The importance of time of dissection was indicated. Midsummer dissections in Ohio give little information on the relation of yield to borers or cavities. For Minnesota differences were observed between the use of borers or cavities for midsummer and fall dissections, cavities consistently appearing as a more stable criterion. The Iowa data revealed that fall dissections of first brood populations affected by natural second brood populations gave almost no correlation between yield and borers but very often correlation of yield with cavities. The value of midsummer dissections in Iowa was indicated.

No differences were observable in the effectiveness of borers or cavities for estimating yield losses in terms of the number of estimating equations obtained or the relative variation of the yield loss-regression coefficients. Cavities were suggested as being a more stable criterion in the presence of interactions between first and second broods.

Regression coefficients for Iowa data were higher for the first brood using midsummer dissections than the second brood. Results indicated that the second brood in Iowa does cause reductions in yield in addition to that resulting from breakage. The regression coefficients for the Ohio data on second brood were generally higher than those for the Iowa data on the second brood.

The resistance of the hybrid was a factor in the success of obtaining a significant regression equation, particularly with first brood populations.

An explanation for the occurrence of positive correlations was made in terms of the interactions of the two broods. The relation between populations of the two broods was found to be important when studying yield losses and suggests the use of all possible infestation level-brood combinations.

Finally, considerable variation from year to year in the estimation of losses indicates the importance and value of long term experiments.

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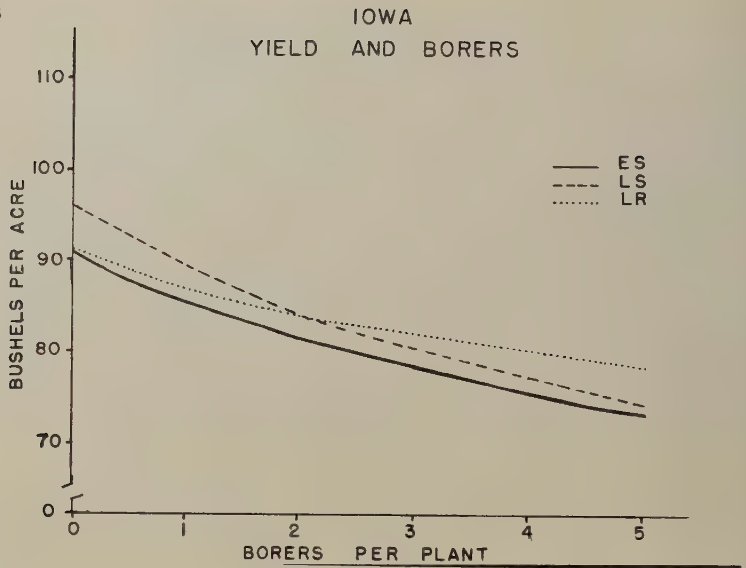

Fig. 1. Pooled regression lines for midsummer dissections in Iowa relating borers per plant per acre to bushels of corn per acre.

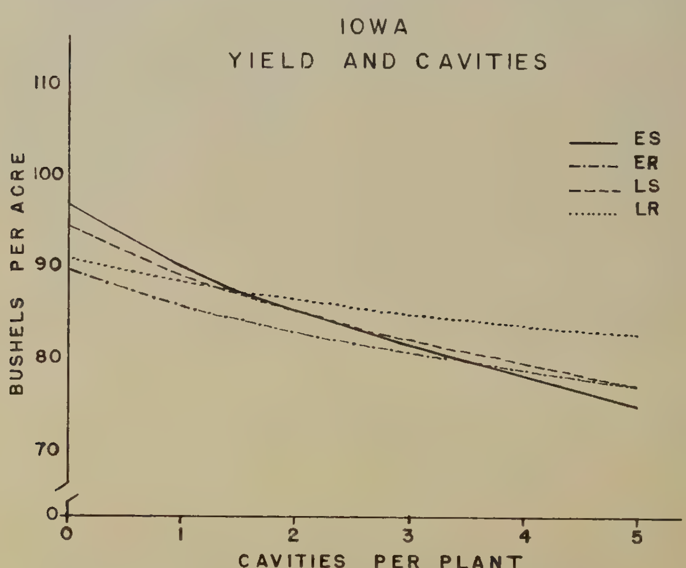

Fig. 2. Pooled regression lines for midsummer dissections in Iowa relating cavities per plant per acre to bushels of corn per acre.

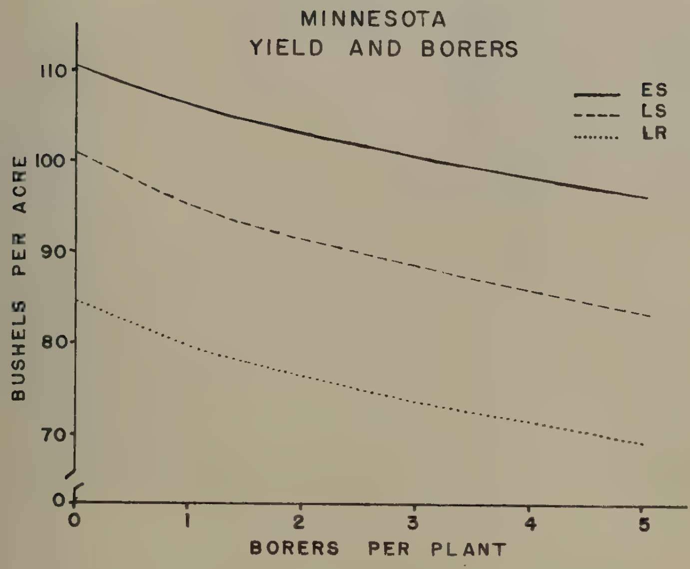

Fig. 3. Pooled regression lines for midsummer dissections in Minnesota relating borers per plant per acre to bushels of corn per acre.

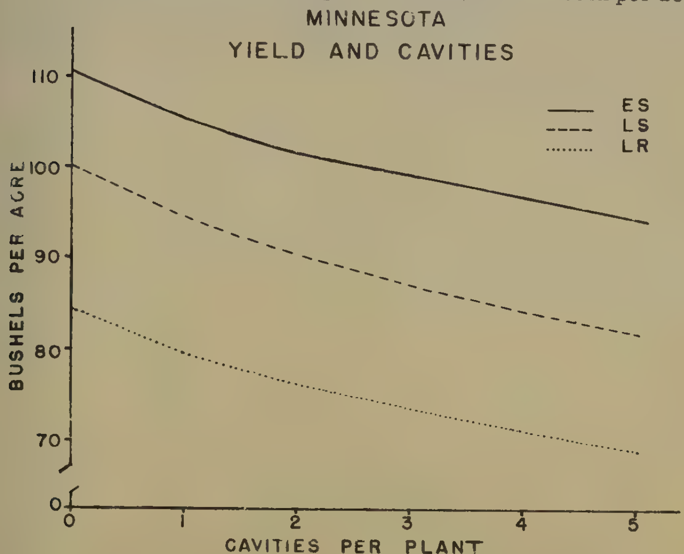

Fig. 4. Pooled regression lines for midsummer dissections in Minnesota relating cavities per plant per acre to bushels of corn per acre.

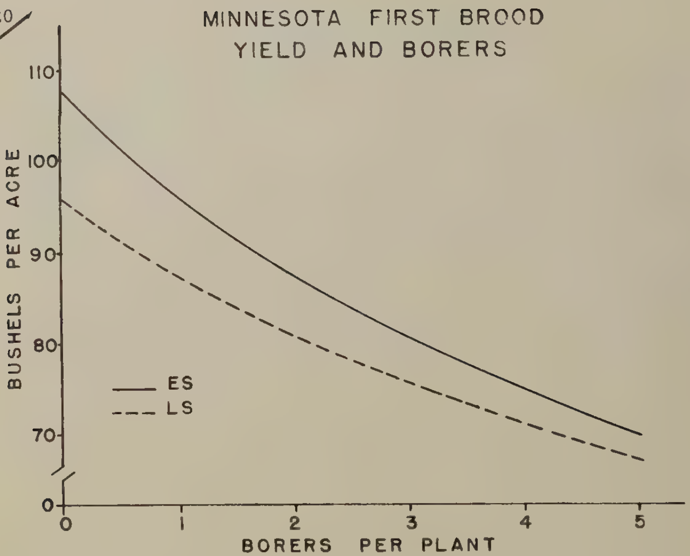

Fig. 5. Pooled regression lines for fall dissections of first brood in Minnesota relating borers per plant per acre to bushels of corn per acre.

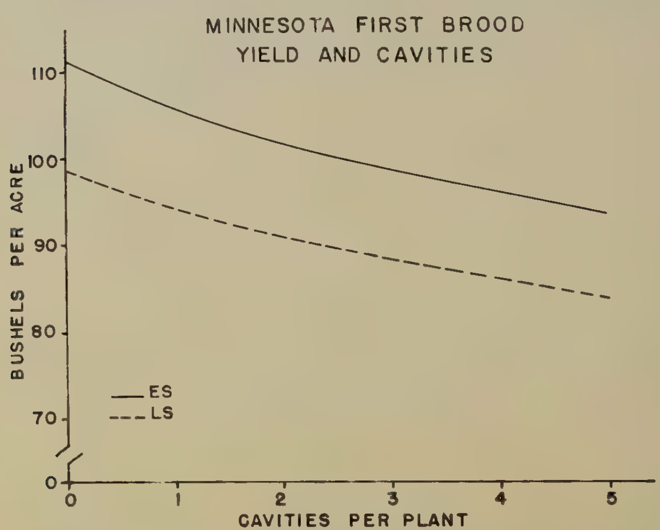

Fig. 6. Pooled regression lines for fall dissections of first brood in Minnesota relating cavities per plant per acre to bushels of corn per acre.

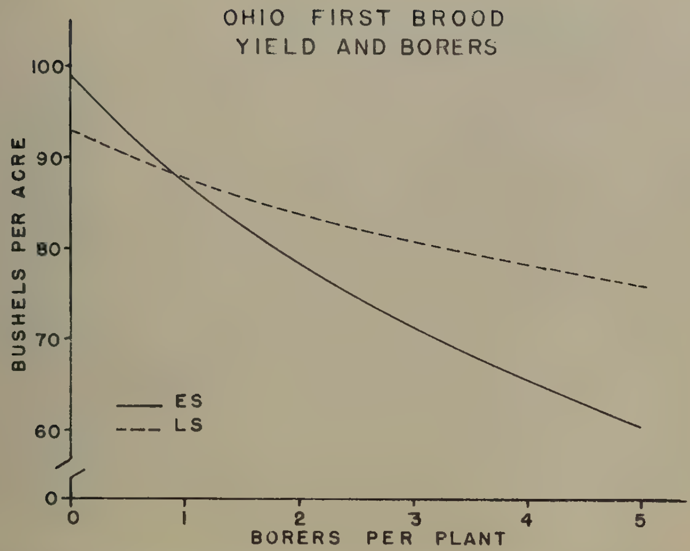

Fig. 7. Pooled regression lines for fall dissections of first brood in Ohio relating borers per plant per acre to bushels of corn per acre.

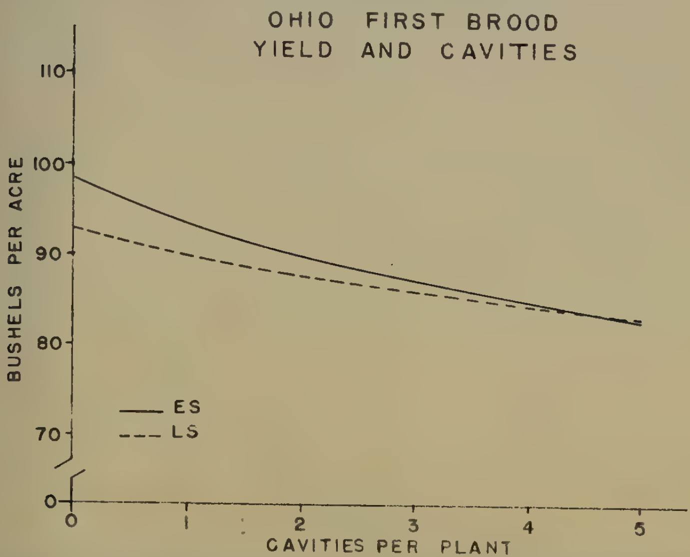

Fig. 8. Pooled regression lines for fall dissections of first brood in Ohio relating cavities per plant per acre to bushels of corn per acre.

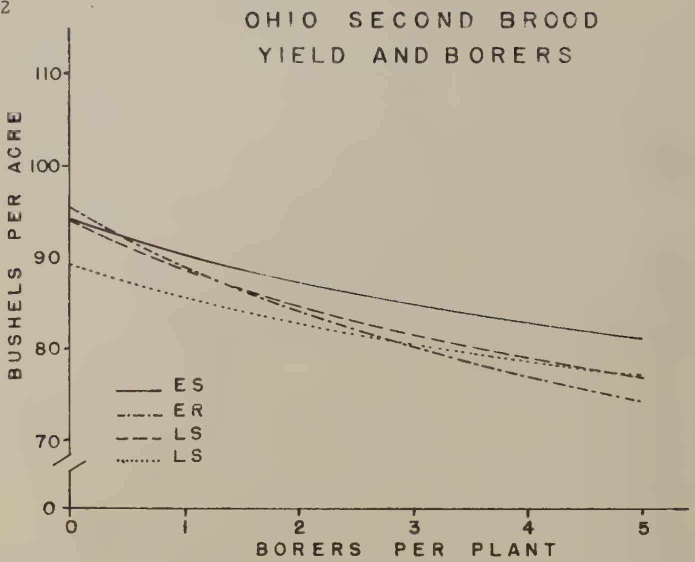

Fig. 9. Pooled regression lines for fall dissections of second brood in Ohio relating borers per plant per acre to bushels of corn per acre.

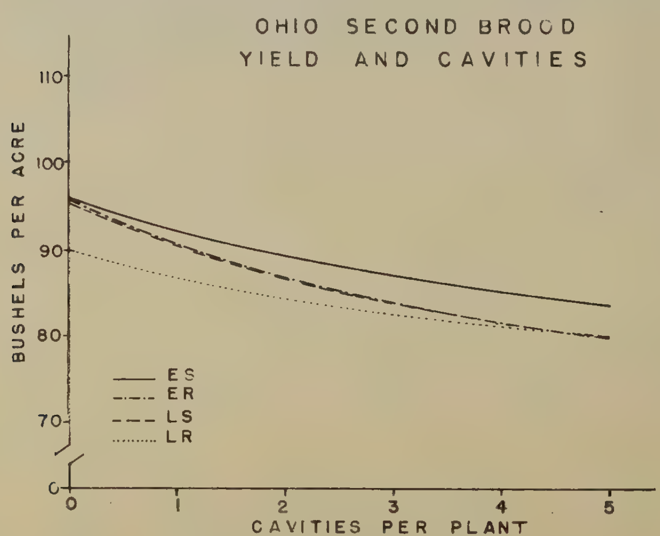

Fig. 10. Pooled regression lines for fall dissections of second brood in Ohio relating cavities per plant per acre to bushels of corn per acre.

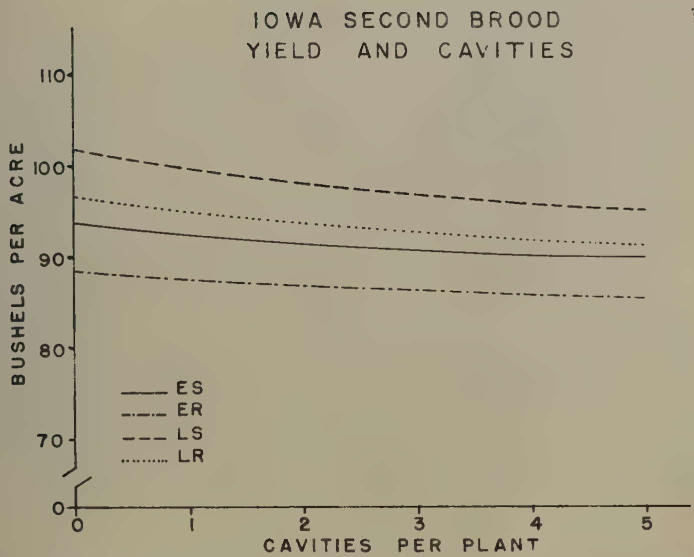

Fig. 11. Pooled regression lines for fall dissections of second brood in Iowa relating borers per plant per acre to bushels of corn per

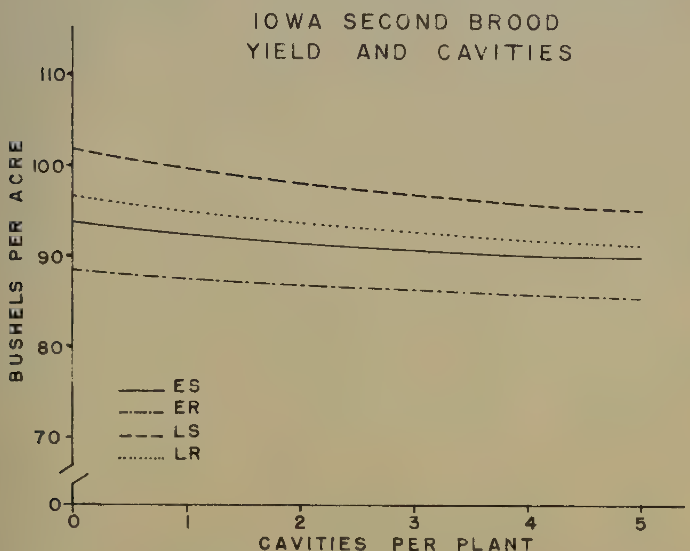

Fig. 12. Pooled regression lines for fall dissections of second brood in Iowa relating cavities per plant per acre to bushels of corn per acre.

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IOWA ASCOMYCETES III. DIAPORTHACEAE : DIAPORTHEAE[1]

Joseph C. Gilman, Lois H. Tiffany, and R. M. Lewis

The section Diaportheae of the Diaporthaceae is the natural continuation of the authors' studies on the Iowa Ascomycetes (Gilman and Tiffany, 1952; Gilman, Tiffany and Lewis, 1957). Like the Valseae, the Diaportheae are predominately found on the stems of flowering plants particularly woody plants with a few more exceptions in the latter on herbaceous plants. Included in the group are species in the following genera: Apioporthe, Cryptodiaporthe, Cryptospora, Cryptosporella, Diaporthe, Diaporthopsis, Endoxyla, Melanconis, Phomatospora, Phragmodiaporthe, Prosthecium and Pseudovalsa.

Taxonomically, the arrangement of genera and species follows closely the dispositions made in this area of mycology by Wehmeyer (1933, 1941). Certain modifications, particularly the inclusion of Endoxyla Fuckel and Phomatospora Saccardo follow the recent treatment of von Arx and Mueller (1954). These authors use the presence of an apical ring in the ascus as an important character of the Diaporthales and merge the Gnomoniaceae with the Diaporthaceae (von Arx, 1951). Until such time that we can make further studies, the species formerly placed in the Gnomoniaceae will not be treated. The family concepts have been reviewed recently (Gilman and Tiffany, 1958) and need no additional discussion in this context. The following genera that are included in the Diaporthaceae by von Arx and Mueller and would be members of the Diaportheae as here interpreted have no Iowa representatives: Heteropera and Mazzantia with their erumpent sclerotial-like stromata and a single perithecium in each stroma in the former; Ditopella and Rehmiella with their 16-spored asci, and clypeus-like stromata, the latter with an extended ostiole, and Gibbelia with its deeply sunken stroma with the parallel dark lines in the substrate and curved necks on the perithecia. Petrak (1955) described Dictyoporthe with muriform ascospores which would be included in this family but as yet no species of the genus have been found in Iowa. The same is true for Titania (Wehmeyer, 1941b) that has a stroma like that of Pseudovalsa but with one-spored asci.

In Iowa the collections of E. W. Holway made in the late 1880's and early 1890's and sent to Ellis have composed the bulk of our known species within the Diaportheae. Most of these were described in Ellis and Everhart's North American Pyrenomycetes (1892). Since that time Gilman and McNew (1940) enumerated the species of Diaporthe, Apioporthe, Cryptodiaporthe and Pseudovalsa from Iowa. They reported 12 species of Diaporthe, 2 of Apioporthe, 2 of Pseudovalsa and 1 of Cryptodiaporthe. Recent collections have markedly expanded these numbers so that in this report 27 species of Diaporthe are listed. In the other genera reported by them, 2 species of Cryptodiaporthe, 2 species of

[1]Journal paper No. J-3523 of the Iowa Agricultural and Home Economics Experiment Station, Ames, Iowa. Project No. 1048.

Pseudovalsa and 2 species of Apioporthe (1 new) have been added. Further additional genera, Cryptospora with 7 species, Cryptosporella with 4 species, Phragmodiaporthe with 1 species, Prosthecium with 3 species, Melanconis with 16 species, Diaporthopsis with 1 species, Phomatospora with 1 species, and Endoxyla with 2 species, have been included. One species of Pseudovalsa, P. ulmi, has been transferred to Prosthecium.

The authors are indebted to Dr. D. P. Rogers for the privilege of examining collections in these genera from the herbarium of the New York Botanical Garden and to Dr. G. W. Martin for the opportunity of studying comparable material from the herbarium of the State University of Iowa. We herewith express our appreciation. Thanks must also be given to Dr. J. A. Stevenson who made the Iowa material in the herbarium of the Office of Mycological Collections at the Bureau of Plant Industry, United States Department of Agriculture, Beltsville, Maryland, available. The source of these specimens is indicated under the respective species by the initials (SUI) for the State University of Iowa and (NYBG) for the New York Botanical Garden. The Iowa material cited in Ellis and Everhart (1892) and Wehmeyer (1933, 1941a, 1941b) are indicated by the inclusion of their names in the list of collections.

A summary of the species, genera and number of host plants follows:

GENUS	NO. OF SPECIES	NO. OF HOSTS
Cryptospora	7	5
Cryptosporella	4	5
Diaporthopsis	1	1
Phomatospora	2	2
Endoxyla	2	1
Apioporthe	4	4
Melanconis	16	17
Diaporthe	27	44
Cryptodiaporthe	3	6
Phragmodiaporthe	1	1
Prosthecium	3	3
Pseudovalsa	3	4

Key to the Genera of the Iowa Diaportheae

a. Ascospores long, fusoid or cylindrical. . . . . . 1. Cryptospora
aa. Ascospores ellipsoid
  b. Ascospores one-celled, hyaline
    c. Stromata present
      d. Stromata valsiform, perithecia circinate with necks converging. . . . . 2. Cryptosporella
      dd. Stromata reduced to a clypeus or a dark line surrounding the entostromata. . . 3. Diaporthopsis
    cc. Stromata lacking
      d. Perithecia small to medium with cylindric necks, asci narrowly cylindric. . . . . 4. Phomatospora
      dd. Perithecia large with tapering necks, asci clavate. . . . . 5. Endoxyla

bb. Ascospores septate
c. Ascospores two-celled
d. Cells of unequal length. . . . . 6. Apioporthe
dd. Cells of equal length
e. With blackened marginal zones
f. With Melanconium conidial stage. . . 7. Melanconis
ff. With Phomopsis conidial stage. . . . 8. Diaporthe
ee. With no blackened marginal zones. . 9. Cryptodiaporthe
cc. Ascospores many-celled
e. Marginal zones prominant. . . . 10. Phragmodiaporthe
ee. Marginal zones absent or not prominent
f. Ascospores appendaged. . . . . 11. Prosthecium
ff. Ascospores not appendaged. . . . 12. Pseudovalsa

1. Cryptospora Tul. pro parte Wehm.

Stromata isolated. Ectostroma forming a small conical disc. Entostroma not developed; perithecia buried in the unaltered bark; no marginal line present. Ascospores long-cylindrical, hyaline, one or more celled or faintly septate.

Conidia elongate-fusoid or cylindrical, borne in open cavities or layers within an ectostroma; sometimes with a second type of smaller, filiform or cylindrical curved, hyaline, one-celled conidia.

The genera Cryptospora, Cryptosporella and Cryptodiaporthe have similar stromatic development. They differ from one another by the ascospore morphology and the associated conidial stages. In Cryptospora the ascospores are elongate to cylindric, one or more celled; in Cryptosporella they are ellipsoid and one-celled while in Cryptodiaporthe they are ellipsoid and two-celled. Karsten in 1873 set up the genus Sillia for the species that were similar to those of Cryptospora but that had multicellular ascospores basing his new genus on Sillia ferruginea (Pers.) Karst. In 1918 von Hoehnel (1918) added Cryptospora albofusca (Cke. and Ell.) Sacc. to the genus. In addition to the multicellular condition of the ascospores, the added character of an erumpent stroma was included. Septation of the ascospores was considered doubtful since von Hoehnel designates them as pseudo-septate. Since this second character, erumpent stroma, is quite variable and present in varying degrees in other members of the genus Cryptospora and since the shape of the spores seemed to show a close relationship of the above two species to other species of Cryptospora, these species are retained in that genus. With this interpretation, the species assigned to these genera have been rearranged.

Key to the Species of the Genus CRYPTOSPORA

a. Ascospores more than 50 microns long
b. Ascospores cylindric
c. Ascospores with rounded ends
d. Ascospores twisted, 48-100 microns long. . 1. C. corylina
dd. Ascospores curved, 50-65 microns long. . 2. C. suffusa
cc. Ascospores with pointed ends. . . . . 3. C. ferruginea

bb. Ascospores thickened at both ends. . . . . 4. C. femoralis
aa. Ascospores less than 50 microns long
b. Ascospores cylindrical
c. Ascospores narrow, 3-4 microns in diameter. . 5. C. betulae
cc. Ascospores thick, 5-6.5 microns in diameter. . 6. C. tiliae
bb. Ascospores subclavate. . . . . 7. C. albofusca

1. Cryptospora corylina (Tul.) Fckl. Figs. 1-5

Stromata in bark, oval or suborbicular, plano-convex, disc conical with raised margin; perithecia 10-25 in each stroma, circinate, minutely globose, slightly decumbent with erect tapering ostioles; ostioles short cylindric, rounded at tip, sometimes dilated; asci obovate or oblong, sessile, 8-spored, 80-120 x 11-15 microns; ascospores twisted or fasciculate, cylindric, one-celled, hyaline, 48-100 x 3.5-5 microns, with many guttulae.

Pycnidial stromata somewhat smaller than the perithecial, obtusely conic with a single locule; conidia linear-cylindric, sometimes slightly curved, 16 x 2-2.5 microns.

On Corylus sp., Decorah, July 6, 1884, E.W. Holway

2. Cryptospora suffusa (Fr.) Ell. and Ev. Figs. 212-215

Stromata scattered, raising the epidermis into flattish pustules, not discoloring it, 1-2.5 mm broad; perithecia 4-12, subcircinate, depressed-globose or angular from crowding, collapsing when dry, lying in the unaltered inner bark, with long, cylindrical converging decumbent necks, united at their ends in a small black erumpent disk which may be obliterated, or sometimes a part or all of them remaining isolated; asci oblong, sessile, eight-spored, 70-100 x 22-30 microns; ascospores curved, fasciculate or interwoven, cylindrical, obtuse, hyaline, 50-65 x 3-4 microns.

On Alnus rugosa (Du Roi) Spreng., Ames, June 24, 1958, L.H. Tiffany

3. Cryptospora ferruginea (Pers.) comb. nov. Figs. 259-262

Sillia ferruginea (Pers.) Karst. Saccardo Syll. 2:361. 1883

Perithecia immersed in an erumpent cushion-like stroma suborbicular to elongate, superficially black, ovoid to globose, monostichous with exserted black ostioles, somewhat flexuous; asci sessile, fusoid-elongate, 80-95 x 12-15 microns, eight-spored; ascospores filiform with pointed ends, somewhat curved, pluri-guttulate or falsely 6-7 septate, hyaline, 60-65 x 3-4 microns.

On Corylus americana Walt., Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany; Pilot Knob State Park, Aug. 6, 1958; L.H. Tiffany

4. Cryptospora femoralis Pk. Figs. 6-8

Pustules small; perithecia few, nestling in inner bark; ostioles few, short, black, erumpent throughout small and mostly transverse chinks in bark, crowded or scattered; asci lanceolate; ascospores crowded, elongate, sublinear, straight or slightly flexuous, obtuse, slightly thickened at the ends, 35-75 microns long. Perithecia adhere to epidermis and are torn off with it.

On Alnus sp., Decorah, May, 1892, E.W. Holway

5. Cryptospora betulae Tul. Figs. 9-12

Perithecia 6-10, globose, small, 0.5 mm or a little larger, circinate, immersed in surface of unaltered inner bark which is raised into slight pustules; necks convergent, decumbent, erumpent in rather prominent, punctiform black ostioles; disc small black, just visible through short transverse cracks in epidermis; asci cylindric-clavate, p. sp. 55-60 x 15-18 microns, aparaphysate; ascospores fasciculate, cylindrical, subarcuate, obtuse, hyaline, 30-40 x 3.5-4 microns.

Conidial stage; Cryptosporium neesii Cda. and C. betulinum Sacc.; conidia 50 x 4.5-5 microns, shaped like ascospores.

On Betula sp., Ames, June 3, 1955, R.M. Lewis

6. Cryptospora tiliae Tul. Figs. 13-15

Pustules irregularly scattered, orbicular, small, slightly prominent, cortical, erumpent disc about 1 mm broad; perithecia 4-5, circinate, subglobose, necks short, decumbent with sub-hemispherical ostioles; asci obovate, 65-80 x 13-16 microns, obtuse above, subacute below; ascospores fasciculate, straight, cylindrical or sometimes subclavate, 30-35 x 5-6.5 microns.

Conidial stage with conidia in an acervulus, fusoid, acute, straight or curved, hyaline, 40-50 x 6.5-10 microns.

On Tilia americana L., Decorah, June 4, 1893, E.W. Holway;
Fort Defiance State Park, Aug. 6, 1958, D. Humphreys;
Fort Defiance State Park, Aug. 20, 1958, L.H. Tiffany

7. Cryptospora albofusca (Cke. and Ell.) Sacc.

Pustules small, covered by slightly raised epidermis, scattered; perithecia few, 4-8 in stroma of scarcely altered substance of inner bark, pale; necks short, small black erumpent ostioles first covered by mealy white pseudo-disc which soon disappears; asci oblong-clavate, sessile, 65-80 x 8-10 microns; ascospores lying parallel in asci, cylindrical or clavate-cylindrical, 30-40 x 3.5-4 microns.

On Quercus sp.

Species not seen

Cryptospora pennsylvanica (B. and C.) Ell. and Ev. was reported from Iowa by Ellis and Everhart (1892) on Prunus americana Marsh. Valsa prunicola Pk. and Diaporthe cylindrospora Pk. are listed as synonyms. Hence it would seem probable that Cryptospora pennsylvanica refers to the specimens listed in this paper as Diaporthe prunicola (Pk.) Wehm.

2. Cryptosporella Sacc.

Stromata isolated. Ectostroma forming small conical disc. Entostroma not developed; perithecia buried in the unaltered bark; no marginal line present. Ascospores elliptical to fusoid, hyaline, one-celled.

Conidia elliptical to fusoid, hyaline, one-celled, produced in more or less open cavities or locules within an ectostroma. Sometimes a second type of smaller, cylindrical or filiform, curved, hyaline, one-celled conidia.

Key to the Species of the Genus CRYPTOSPORELLA

a. Perithecia mostly one in a pustule; ascospores cylindrical, 2 microns in diameter. . . . . 1. C. lentaginis
aa. Perithecia several in each pustule; ascospores oblong-elliptical to subelliptical
b. Ascospores 30-60 x 7-10 microns. . . . . . . 2. C. hypodermia
bb. Ascospores 11-15 microns long
c. Ascospores oblong elliptical, 11-13 x 4-4.5 microns. . . . . 3. C. punctostoma
cc. Ascospores subelliptical, 11-15 x 4-6 microns. 4. C. viticola

1. Cryptosporella lentaginis Rehm

Pustules 1 mm diameter; perithecia mostly one in a pustule, 0.5 mm diameter, obtuse-conical from a globose base, contracted above into stout, obtusely conical ostiole which barely pierces slightly elevated epidermis; asci clavate, stipitate, 40-50 x 8-9 microns; ascospores biseriate, cylindrical, curved or nearly straight, continuous, 2-3 guttulate, hyaline, 12 x 2 microns.

Inner bark is uniformly blackened, the numerous small stromata are light colored within and appear as light colored spots in a longitudinal section through the bark. The species is anomalous by having for the most part a single perithecium in a stroma.

On dead Viburnum lentago L., Cedar Falls, Jessie A. Parrish (SUI); Decorah, June, 1882, E.W. Holway (NYBG); Ellis and Everhart (1892)

2. Cryptosporella hypodermia (Fr.) Sacc. Figs. 263-266

Perithecia united into valsa-like groups of three to ten, erumpent from the swollen pustules by cylindrical, elongate convergent necks, globose to irregular, 400-700 microns in diameter, nesting in the bark parenchyma, necks 400-650 x 170-220 microns; asci spindle- to club-shaped, truncate above, narrowed below with a delicate simple wall weakly thickened at the tip, 90-120 x 15-20 microns, eight-spored; ascospores two or three ranked, ellipsoid-spindle-form, one-celled, hyaline, 30-60 x 7-10 microns, paraphyses not observed.

On Acer saccarinum L., Lost Island State Park, Aug. 13, 1958, L.H. Tiffany

3. Cryptosporella punctostoma (Ell.) Wehm. Figs. 16-18

Stroma cortical, of unaltered substance of innerbark; perithecia 8-12, 0.5 mm diameter, in a single layer; short-cylindrical beaks joined in small olivaceous, slightly elevated disc, ostioles erumpent circinately; asci clavate-cylindrical, 55 x 8-9 microns; ascospores biseriate, oblong-elliptical, 4-guttulate, slightly constricted in middle, hyaline, 11-13 x 4-4.5 microns.

On Amelanchier canadensis (L.) Medic, Decorah, July 1, 1882, E.W. Holway (type); Ellis and Everhart (1892) as Diaporthe stictostoma (Ell.) Sacc.; Wehmeyer (1933)

Ellis (1883b) described this species under the name Valsa punctostoma. The species was transferred into Cryptosporella by Wehmeyer (1933).

4. Cryptosporella viticola Shear Figs. 19-22

Perithecia buried in irregularly pulvinate subcorticular stromata, thin-walled, globose, with short stout slightly exserted beak; asci sessile or subsessile, 60-72 x 7-8 microns; paraphyses slender, septate, exceeding asci in length; ascospores subelliptic, obtuse, hyaline, continuous, 11-15 x 4-6 microns.

Pycnidia very variable in size, structure and arrangement; usually with irregular labyrinthiform chambers, ostiolate, but frequently rupturing and upper half of pycnidium falling away; conidia of two kinds, one hyaline, nonseptate, subfusoid, straight or slightly curved, 7.5-15 x 2-5 microns, others scolecosporous, hyaline, long slender, nonseptate, variously curved or hooked, 18-30 x 1.0-1.5 microns.

On Parthenocissus sp., Ames, May 8, 1955, R.M. Lewis

On Vitis sp. (cult.), Logan, H.E. Nichols, det. W.A. Archer

We have not seen the ascus stages of this species. The conidial stage is rather common on both grape and Virginia creeper.

3. Diaporthopsis Fabre

Perithecia immersed, scattered singly or crowded within an evenly effuse entostromatic area, separately erumpent. Surface of substratum usually more or less blackened. Entostromatic areas often margined by a blackened zone. Asci clavate, with evanescent stalks which free the asci within the perithecium. Spores hyaline, one-celled, ellipsoid to fusoid, straight or curved, sometimes appendaged.

1. Diaporthopsis angelicae (Berk.) Wehm. Figs. 23-26

Surface of substratum becoming more or less blackened over wide areas at maturity, exposed areas often show a violet tinge, ventral zones absent or rather faintly developed within the pith; ostioles small, barely erumpent as small papillate to cylindric or conic projections; perithecia small, spherical or flattened, 240-400 x 160-240 microns, scattered singly or sometimes crowded, usually rather deeply immersed within the pith and with long slender necks, broad paraphysis-like hyphae present in young perithecia; asci clavate at first, becoming long-cylindric, 40-50 x 4-7 microns; ascospores biseriate to obliquely uniseriate, fusoid-ellipsoid, one-celled, hyaline, not constricted, 9-15 x 3-4 microns.

Phomopsis conidial stage; alpha conidia ellipsoid to oblong, 5-8 x 2-2.5 microns; beta conidia oblong-cylindric, 10 x 3 microns.

On overwintered herbaceous stems, Ames, Sept. 24, 1957,
L.H. Tiffany; Marble Lake, Aug. 11, 1958, L.H. Tiffany

This species might be confused with Diaporthe arctii. It is most clearly separated by the reddish to violet tinge of the stroma, the irregular and indefinitely delimited stroma, smaller perithecia which are usually deeply imbedded, and the ascospores which are three-guttulate with a central guttula.

4. Phomatospora Sacc.

Saprophytes with sunken, small to medium-sized globose perithecia, erumpent by a short cylindrical to papillate mouth pierced by a canal lined with periphyses; peridium membranous with an outer layer of angular rather thin-walled, brown cells and an inner hyaline sheath; asci

cylindrical, stipitate, with delicate, thin walls and a small difficultly observed apical ring; ascospores 8, usually one-celled, biseriate; paraphyses thin-walled, dissolving early.

Key to the Species of the Genus PHOMATOSPORA

a. Ascospores 18-20 x 8-9 microns. . . . . . . 1. P. botrysphaerioides
aa. Ascospores 6-10 x 2-3 microns. . . . . 2. P. berkeleyi

1. Phomatospora botryosphaerioides Speg.
Perithecia subepidermal, gregarious, discrete, conoidal-lenticular, 200-250 microns in diameter, membranous, piercing the epidermis by a papillate ostiole, black, smooth, subcarbonaceous; asci cylindrical, rounded at the apex with an indistinct thickened ring, with a short stipe, 85-90 x 18 microns, 8 spored, aparaphysate; ascospores biseriate, elliptical or subrhomboid, 18-20 x 8-9 microns, one-celled, hyaline.
On dead culms of Scirpus sp., Lake Okoboji, Iowa, August, 1956, L.H. Tiffany

2. Phomatospora berkeleyi Sacc. Figs. 267-270
Perithecia scattered, deeply sunken in the substrate, globose, membranous, 140-210 microns in diameter, erumpent through the covering membrane by a short cylindrical neck of 40-70 x 40-55 microns; asci cylindrical, standing parallel with one another, abruptly rounded at the tip, 60-90 x 3.4-6 microns, eight-spored; ascospores uniseriate, one-celled, hyaline, 6-10 x 2-3 microns; paraphyses delicate, thin-walled, thread-like.
On herbaceous stem, Marble Lake, Aug. 11, 1958, L.H. Tiffany

5. Endoxyla Fuckel
Perithecia sunken in the substrate, becoming exposed by weathering, rather large, globose or flattened by pressure, with long apical, or at times sublateral, cylindric necks, clothed with periphyses; asci cylindric to subclavate, standing parallel over entire inner surface of the perithecia, with an apical ring, eight-spored, sessile; ascospores ellipsoid, hyaline to smoky; usually one-celled, but may become divided in age into two, four, or eight cells by cross-walls.
As here described, Endoxyla contains the species of Ceratostomella that have persistent asci and an apical refractive ring.

Key to the Species of the Genus ENDOXYLA

a. Ascospores cylindric with rounded ends, becoming 3-septate, 6-13 x 3-4.5 microns. . . . . 1. E. cirrhosa
aa. Ascospores ellipsoid one-celled, 10-15 x 3-5 microns. 2. E. operculata

1. Endoxyla cirrhosa (Pers.) Arx and Mueller Figs. 32-35
Perithecia sunken in substrate, black, globose, 280-500 microns with apical or at times slightly lateral short cylindric or elongate necks 200-1200 microns long; at times perithecia covered with red-brown hyphae,

walls tough leathery to carbonaceous; asci more or less parallel covering the interior walls, cylindrical with stipe 20-40 microns long, sporiferous part 50-80 microns; strong refractive apical ring; paraphyses sparse; ascospores ellipsoid, uniseriate, one-celled, hyaline at first, becoming one to three septate, 6-13 x 3.0-4.5 microns.

On decaying wood, Amana, October 5, 1957, L.H. Tiffany;
Lake Okoboji, August, 1956, L.H. Tiffany

2. Endoxyla operculata (Alb. and Schw.) Fckl. Figs. 27-31

Perithecia scattered, singly or in groups, often sunken in substrate, then exposed by weathering and becoming superficial, globose, brownish-black, 300-500 microns in diameter, covered at the base with a tomentum of red-brown septate hyphae, 3-5 microns in diameter; ostioles black, slightly tapered neck, 300-1500 microns long; asci cylindric, standing in a hymenium, uniseriate, 55-80 x 6-8 microns, with a refractive apical ring; ascospores ellipsoid with rounded ends, one-celled, slightly smoke-brown, 10-15 x 3-5 microns. In young perithecia paraphyses are numerous.

On wood, Ames, 1957, L.H. Tiffany

6. Apioporthe (Hoehn.) Wehm.

Perithecia immersed in the substratum, usually clustered. Entostromatic development scanty or variously developed as a mycelial weft or definite stromatic tissue about the perithecia. Entostromata also various. Tissues above the perithecia sometimes blackened, but no definite marginal zones within the substratum as Diaporthe. Asci clavate, with a refractive ring in the apex, stalks evanescent. Ascospores hyaline, unequally two-celled, fusoid to pyriform, commonly tapered toward one end, which contains the smaller cell.

Conidial stage consisting of variously shaped cavities formed within a stromatic pycnidial tissue by the simultaneous breaking up of the hyphae in these locular areas, into conidia without the formation of a definite hymenium.

Key to the Species of the Genus APIOPORTHE

a. Ascospores clearly two-celled, cells unequal
  b. Ascospores 11-14 x 2.5-5.5 microns
    c. Ostioles erumpent through orange-colored papillae. . . . . 1. A. corni
    cc. Ostioles erumpent through blackened disk. . 2. A. apiospora
  bb. Ascospores 30-36 x 9-10.5 microns. . . . . . 3. A. macrospora
aa. Ascospores with a caplike second cell. . . . . . . 4. A. anomala

1. Apioporthe corni Wehm.

On surface as minute scarcely visible, papillate, orange-colored pustules, through which one to several ostioles are erumpent; perithecia 200-320 x 160-240 microns, one or several clustered beneath the minute orange-colored ectostromatic disk, and usually embedded in yellowish entostroma; no blackened zones within the substratum; asci long clavate,

54-65 x 5.5-6.5 microns; ascospores biseriate, variable in shape, ellipsoid or inequilateral-fusoid to pyriform, often tapered toward one end, hyaline, tardily becoming unequally two-celled, 12-15 x 2.5-4 microns.

On Cornus sp., Woodman Hollow State Park, Oct. 5, 1958, L.H. Tiffany

2. Apioporthe apiospora (Ell. and Holw.) Wehm. Figs. 40-43

Barely visible on surface as minute pustules with small central blackened disk composed of cluster of few barely erumpent ostioles; no blackened zones within substratum; perithecia 240-320 microns in diameter; in small groups in surface layers of bark, walls thick, membranous, black, walls of several perithecia often fused together to form stroma-like structure; ostioles collectively erumpent; asci 75-80 x 9-10 microns; ascospores obliquely uniseriate to subbiseriate, ellipsoid-ovoid, narrower toward one end, unequally two-celled, hyaline, constricted at septum, 11-14 x 2.5-5.5 microns.

On Ulmus americana L., Ames, Jan. 30, 1958, L.H. Tiffany; Ames, March 23, 1958, L.H. Tiffany

On Ulmus sp., Decorah, May, 1892, E.W. Holway (Diaporthe apiospora Ell. and Holw.); Wehmeyer (1933)

3. Apioporthe macrospora sp. nov. Figs. 255-258

Perithecia in scattered minute papillate pustules under the epidermis, immersed in the brown entostroma below the ectostroma, without evident zones, erumpent singly or in groups by short ostiolar necks; asci long-clavate, 65-77 x 10-12 microns with a refractive apical ring; ascospores biseriate, unequal-sided fusiform or pyriform, often narrowed toward the apex, hyaline unequally bicellular, 30-36 x 9-10.5 microns.

On Quercus alba L., Fort Defiance State Park, Aug. 7, 1958, L.H. Tiffany

Apioporthe macrospora sp. nov.

Sori in aspectu superficiali minuti papillati uno vel pluribus ostioles erumpentibus praediti. Perithecia 300-480 x 160-240 μ, aggregatim vel solitarie sub ectostroma brunneum minutum in entostromate immersa. Zonae nigricantes desunt. Asci longi-clavati 65-77 x 10-12 μ, cum cingulo refractivo ad apicem. Ascosporae biseriatae, inaequilateraliter fusiformes vel pyriformes, saepe apicem versus angustatae, hyalinae, inaequiliter bicellulae, 30-36 x 9-10.5 μ.

In ramis corticates emortuis Quercinis albae.

4. Apioporthe anomala (Pk.) Hoehn. Figs. 36-39

Pustules prominent, longitudinally elongate blackish discs, rupturing the periderm, 2-5 mm in length and about 2 mm in diameter, and showing the blackened circular, slightly sulcate ostioles; perithecia elongate, crowded in the blackened stroma and reaching to the wood; asci clavate, 33-42 x 10-12 microns; ascospores biseriate, ellipsoid with a somewhat pointed end which is composed of a minute caplike second cell, 10-12 x 4 microns.

On living Corylus americana Walt., Decorah, September, 1882, E.W. Holway. Compare Ell. and Ev. N.A.F. 1185.

On dead Corylus sp., Decorah, Oct., 1882, E. W. Holway (NYBG)

Ellis and Everhart (1892) reported this fungus from Iowa as Cryptospora anomala (Pk.) Ell. and Ev.

7. Melanconis Tul. emend. Wehm.

Stromata isolated. Disc consisting of a well-developed conical ectostroma. Perithecia arranged circinately in the unaltered bark tissue. No entostromatic development apparent, and no dark zone present. Paraphyses present, sometimes fairly numerous and persistent. Ascospores biseriate to uniseriate, elliptical, two-celled, hyaline or brown, sometimes appendaged.

Conidial stage belonging to the form genus Melanconium. Hymenial layers formed over the entire surface, or within cavities upon the flanks of a conical or pulvinate ectostroma. Conidia of two types, at least in some species; one, small, cylindrical to elliptical, one-celled, and hyaline; the second, larger, elliptical to fusoid, one-celled and colored.

Key to the Species of the Genus MELANCONIS

a. Ascospores hyaline
  b. Ascospores appendaged
    c. Appendages as long or longer than ascospores
      d. Appendages strongly curved. . . . . 1. M. occulta
      dd. Appendages not strongly curved
        e. Spores strongly curved. . . . . 2. M. thelebola
        ee. Spores slightly curved. . . . . 3. M. everhartii
    cc. Appendages short, caplike or spinelike
      d. Ascospores 10-20 x 3.5-5 microns
        e. Conidia hyaline. . . . . 4. M. chrysostroma var. ellisii
        ee. Conidia of at least one type brown. . . 5. M. ostryae
      dd. Ascospores 20-24 x 8 microns. . . . . 6. M. flavovirens
  bb. Ascospores not appendaged
    c. Ectostroma white to yellowish. . . . . . 7. M. stilbostoma
    cc. Ectostroma gray-green to olive or yellow-green
      d. Ascospores 3.5-7 microns in diameter
        e. Conidia hyaline. . . . . 4. M. chrysostroma var. ellisii
        ee. Conidia of at least one type brown. . . 5. M. ostryae
      dd. Ascospores 6-12 microns in diameter
        e. Melanconium stage unknown. . . 5. M. flavovirens
        ee. Melanconium stage often present
          f. Ascospores 6.5-9 microns in diameter. . . . . 8. M. juglandis
          ff. Ascospores 8-12 microns in diameter. . . . . 8a. M. juglandis var. tiliae
aa. Ascospores brown
  b. Ectostroma white to yellowish. . . . . 9. M. apocrypta
  bb. Ectostroma dark-colored, gray to brown to black

c. Perithecia immersed in the entostroma which is fused with the ectostroma
d. Ascospores 23-27 x 10-13 microns. . . . . 10. M. modonia
dd. Ascospores 15-21 x 7-8 microns. . . . . . 11. M. smilacis
cc. Ectostroma distinct
d. Ascospores with appendages
e. Ascospores 25-38 x 12-15 microns. . 12. M. appendiculata
ee. Ascospores 35-60 x 16-20 microns. . . 13. M. acrocystis
dd. Ascospores without appendages
e. Ascospores less than 30 microns long
f. Ectostroma well developed. . . . 14. M. decorahensis
ff. Ectostroma poorly developed. . . . . . 15. M. corni
ee. Ascospores more than 30 microns long. . . 16. M. sudans

1. Melanconis occulta (Fckl.) Sacc. Figs. 59-61

On the surface as numerous angular to elongate pustulate ruptures 0.3-1.5 mm long, with adherent periderm; ostioles stout hemispheric, scattered or clustered, often united into a black carbonaceous erumpent disc; perithecia single or in small irregular groups, 400-720 x 350-550 microns; thick walls of large brown cells; asci large, stout-clavate, with a thickened apical wall, 100-150 x 25-40 microns; ascospores biseriate to triseriate, large, ellipsoid to cylindric-ellipsoid, ends rounded, two-celled, hyaline, not constricted at the septum, with rather thick walls and one or two guttulae in each cell, 36-50 x 11-20 microns and often a broad hyaline recurved appendage 10-26 x 5-7 microns at each end; appendages more or less evanescent and often absent.

Wehmeyer (1941b) reported no conidial stage produced in cultures and none consistently associated with natural material.

On Populus tremuloides Michx., Fort Dodge, June 15, 1956, L.H. Tiffany

2. Melanconis thelebola (Fr.) Sacc. Figs. 80-83

On the surface as scattered conic pustules 1-2.5 mm in diameter, with central circular or fusoid white discs 0.5-1.5 mm in diameter, barely erumpent through the closely adherent periderm and containing from two to twelve short stout or conic black ostioles; ectostroma yellowish white, sometimes well developed and distinct, more often slight or obliterated and merging with a yellowish entostromatic development in the bark beneath; perithecia 500-800 microns in diameter, more or less circinate with convergent necks, usually in a pustulate swollen area of the bark, may or may not have a blackened zone on the surface, or dipping into the bark cortex; blackened zone sometimes shows through the periderm as a marginal line; asci clavate, 120-195 x 15-21 microns; ascospores irregularly biseriate, ellipsoid, cylindric, straight or more often slightly curved, a little or not at all constricted at the septum, two-celled, hyaline, 24-35 x 8-10 microns, sometimes up to 42 microns long but usually similar in diameter, long hyaline bristle-like appendages at each end when fresh, evanescent in old material; ascospores may become two- or three-septate and brown in age.

Pycnidia with a flattened or irregular cavity in the base of a conic ectostroma with an elongate ostiole obtained in culture by Wehmeyer

(1941b); alpha conidia ellipsoid to oblong, often bent or curved, tapered toward one end, brown, two- to four-celled, 18-40 x 5-12 microns; beta conidia cylindric to inequilateral or tapered, often curved, one-celled, hyaline, 5-9 x 1-1.5 microns.

On Alnus sp., Decorah, Oct. 28, 1894, E.W. Holway

3. Melanconis everhartii Ell. Figs. 52-55

On the surface as pulvinate circular pustules or thickly scattered angular to elongate ruptures of the periderm 0.3-2 x 0.3-1 mm, often arranged in longitudinal series; small cortical disc or loose fascicle of from one to eight black often elongate ostioles; slight development of light ectostroma, soon obliterated by erumpent ostioles; perithecia 200-400 microns in diameter, loosely grouped in upper layers of bark cortex, often causing swollen areas on surface; asci broad-clavate, 80-142 x 20-30 microns; ascospores biseriate to triseriate, fusoid-ellipsoid to cylindric ellipsoid, two-celled, hyaline, coarsely granular, straight or slightly curved, ends rounded, slightly constricted at the septum, 26-64 x 8.5-11 microns, stout filiform slight curved hyaline appendage, 20-25 x 3-4 microns at each end, appendages evanescent.

On Acer saccharinum L., Boone County, May 11, 1958, L. H. Tiffany; Wanata State Park, Aug. 18, 1958, L.H. Tiffany

On Acer sp., Iowa, T.H. MacBride (SUI)

4. Melanconis chrysostroma (Fr.) Tull. var. ellisii Wehm. Figs. 56-58

Small conic pustules with central grayish to olive-green disc 0.2-0.8 mm in diameter, ostioles black, papillate, barely reumpent, scattered throughout or arranged about the margin of the disc; disc often surrounded by circular swollen area caused by the perithecia; perithecia spherical or flattened, 250-500 x 250-480 microns, circinately arranged or clustered; asci clavate, 50-65 x 6.5-13 microns; ascospores biseriate to uniseriate or obliquely uniseriate, fusoid-ellipsoid, two-celled, hyaline, more or less constricted at the septum, ends acute to obtuse, caplike or spinelike evanescent appendages, walls sometimes appearing gelatinous and slightly swollen, 11-20 x 3.5-5 microns.

Melanconium conidial stage; conidia exuded at sides of central disc as pinkish to yellow spore masses, drying to olive-brown; alpha conidia cylindric-fusoid to ellipsoid-fusoid, straight, curved or bent, one-celled, hyaline, 16.5-30 x 3.5-5 microns; beta conidia cylindric, inequilateral or allantoid, hyaline, one-celled, 8-12 x 1.5-3 microns.

On Carpinus caroliniana Walt., Iowa, in Ellis and Everhart (1892) (as Melanconis bitorulosa Ell. and Ev.) on Carpinus americana; Woodman Hollow State Park, Oct. 5, 1958, L.H. Tiffany

On Carpinus sp., Decorah, July 2, 1882, E.W. Holway

On Ostrya virginiana (Mill.) K. Koch, Ames, Sept. 1955, R.M. Lewis; Iowa, in Ellis and Everhart (1892) (as Melanconis bitorulosa Ell. and Ev.)

Holway's collection on Carpinus sp. was originally identified as Diaporthe carpinicola Fckl. and was reported as Melanconis hyperopta (Nit.) Wehm. by Gilman and McNew (1940). Wehmeyer (1941) later considered it as an American variety of Melanconis chrysostroma.

5. Melanconis ostryae (Dearn.) Wehm. Figs. 76-79

On the surface as minute flattened conic pustules with a minute central gray to greenish-gray disc 0.2-0.4 mm in diameter through which few minute black ostioles are barely erumpent; perithecia 300-500 microns in diameter, occurring circinately in small groups in the unaltered bark cortex beneath the small rather distinct gray-green conic ectostroma; asci clavate, refractive ring in apex, 60-85 x 5-8 microns; ascospores irregularly biseriate, fusoid-ellipsoid, two-celled, hyaline, constricted at the septum, often somewhat tapered toward one end and commonly with one cell somewhat slightly smaller, 14-20 x 4-7 microns.

Melanconium conidial stage obtained in culture by Wehmeyer (1941b). Conidia exuded in black spore mass, alpha conidia oblong-ellipsoid to ovoid, one-celled, olive-brown, coarsely granular, with lighter equatorial band, 8.5-14 x 6-7.5 microns; beta conidia rare, elongate-fusoid, one-celled, hyaline, bent or curved, 19.5-25 x 2.5-3.5 microns.

On Ostrya virginiana (Mill.) K. Koch, Iowa

6. Melanconis flavovirens (Otth) Wehm. Figs. 84-87

Stromata on the surface as conically pustulate or circular swellings of the periderm with a central circular to laterally elongate fusoid blackish or dark olive-green erumpent disc through which stout cylindric punctate ostioles emerge; perithecia more or less flattened-spherical, 300-600 x 450 microns, circinately arranged beneath ectostromata; asci clavate, 85-110 x 10-15 microns; ascospores biseriate, two-celled, hyaline fusoid-ellipsoid with rather acute tips, often somewhat inequilateral, slightly constricted at the septum, contents granular to guttulate, sometimes with faint gelatinous, knob-like appendage at each end, walls appearing thickened, 17.5-27 x 6-9.5 microns.

Myxosporium or Discosporium conidial stage; conidia long-fusiform, sometimes inequilateral, continuous, one- to two-guttulate, hyaline, 12-16 x 5-6 microns.

On Corylus americana Walt., Decorah, June, 1892, E.W. Holway (Ell. Fung. Columb. 30), Wehmeyer (1941)

On dead Corylus americana, Decorah, June, 1892, E.W. Holway, N.A.F. 2742 (NYBG)

Labels on N.A.F. 2742 and Ell. Fung. Columb. 30 give Carpinus americana as host. This has been interpreted to be a misprint for Corylus (Wehmeyer, 1941).

7. Melanconis stilbostoma (Fr.) Tul. Figs. 88-93

Rupturing the periderm as numerous circular conic pustules 1-3 mm in diameter with a central white to yellowish disc 0.5-1 mm in diameter through which short stout cylindric ostioles are collectively erumpent often marginally between ectostroma and the closely adherent periderm; perithecia spheric to flattened, 300-500 x 250-350 microns, circinately arranged in the unaltered cortex beneath the well-developed truncate-conic white to yellowish ectostroma often causing swollen prominences around the central pustule; asci clavate-fusoid, with refractive ring, 60-110 x 10-16 microns; ascospores irregularly biseriate, rather broad, ovoid-ellipsoid, ends somewhat tapered at first but becoming rounded, two-celled, hyaline, constricted at the septum when mature and turgid,

often not in young or dried material, coarsely granular, 13-23 x 5-9 microns.

Melanconium conidial stage obtained in culture by Wehmeyer (1941b); alpha conidia ellipsoid to fusoid or ovate, one-celled, brown, 10-21 x 5.5-8.5 microns; beta conidia cylindric, straight or somewhat curved, one-celled, hyaline, 6.5-12 x 2-2.5 microns.

On Betula sp., Decorah, June, 1883, E.W. Holway (NYBG);
(ISC); Ellis and Everhart (1892)

8. Melanconis juglandis (Ell. and Ev.) Graves Figs. 94-96

On the surface as scattered conic pustules about 1 mm in diameter, with central angular gray to gray-green stromatic disc 0.2-0.5 mm in diameter; ostioles stout-cylindric to conic, occasionally elongate, erumpent through the ectostroma; ectostroma usually small but well defined, gray-green to yellow-green on the bark surface; perithecia 400-700 microns in diameter, circinate in the unaltered bark cortex beneath the ectostroma and erumpent through it; asci cylindric-clavate to cylindric, 87-130 x 12-17 microns; ascospores obliquely uniseriate to biseriate, broad, fusoid-ellipsoid, two-celled, hyaline, constricted at the septum, large guttula in each cell, 16-23 x 6.5-9.5 microns.

Melanconium conidial stage; conidia brown, oblong-ellipsoid to irregular, blunt at one end and tapering to a flat apiculate base, 13-25 x 8-10 microns.

On Juglans cinerea L., Decorah, May 9, 1882, E.W. Holway
On Juglans nigra L., Ames, April 4, 1931, J.C. Gilman

8a. Melanconis juglandis var. tiliae Wehm. Figs. 193-196

Pustules slightly more thickly scattered; asci 90-145 x 10-16.5 microns; ascospores 16-22 x 8-12 microns.

Melanconium conidial stage found associated with perithecial pustules by Wehmeyer (1941b); conidia ovoid, pip-shaped with flattened apiculate base, one-celled, brown, 16.5-21.5 x 9-10 microns.

On Tilia americana L., Ledges State Park, July 23, 1957,
L.H. Tiffany
On Tilia sp., Decorah, May 30, 1892, E.W. Holway,
Ell. and Ev. 2826 (NYBG)

9. Melanconis apocrypta Ell. Figs. 223-226

On surface as minute apapillate circular to elliptic swellings, 0.2-0.8 mm in diameter, occasionally faintly outlined by marginal line; pustules finally with minute central rupture through which few minute black ostioles are barely erumpent; perithecia 300-400 microns in diameter, circinate or irregularly crowded in unaltered bark cortex, collectively erumpent by rather elongate necks; asci cylindric-clavate becoming cylindric, 150-200 x 18-26 microns; ascospores irregularly biseriate, becoming uniseriate, broad-ellipsoid, two-celled, constricted at septum, contents granular, soon with single large guttula in each cell, hyaline, becoming dark brown, 23-32 x 12-14 microns with an evanescent gelatinous envelope. Surface of pustules may be blackened by exuded ascospores.

Melanconium conidial stage; conidia oblong-ellipsoid, one-celled, 13-16.5 x 5.5-6.5 microns; conidia obtained by Wehmeyer (1941b) in culture were ovoid to fusoid-ellipsoid, brown, mostly one-celled, occasionally two-celled, 15-45 x 8.5-11.5 microns.

On Populus sp., Decorah, July 14, 1882, E.W. Holway, No. 164 Ell. Coll. (ISC) (NYBG) (type); Ellis and Everhart (1892); Wehmeyer (1941b)

10. Melanconis modonia Tul. Figs. 62-65

Circular to fusoid flat pulvinate pustules 1-2 mm in diameter, with closely adherent periderm and central slit-like or angular rupture exposing a gray-black carbonaceous disc 0.2-1 mm in diameter, ostioles barely erumpent or not visible, ectostroma conic to truncate-conic, greenish-brown to brown, fused with entostroma; perithecia spheric to elongate, 350-700 microns in diameter, in small groups within a differentiated entostromatic area of bark penetrated by interwoven brownish hyphae which give a greenish-black to brownish discoloration; asci cylindric-clavate to cylindric with thickened apical wall, 120-200 x 11-25 μ; ascospores biseriate to irregularly uniseriate, ellipsoid-fusoid, two-celled, hyaline becoming brownish when fully mature or when exuded onto twig, constricted at septum, ends rounded, 23-37 x 10-13.5 μ.

Conidia borne in circular flattened pustules with a central pore or angular rupture, conidia ovate-lanceolate, pyriform to clavate, straight to curved, two- to eight-celled, brown, somewhat constricted at septa, large guttula in each cell, 20-74 x 8-14 microns, often with remains of stipe cell at base.

On Castanea dentata (Marsh.) Borkh., Iowa, CC No. 298

11. Melanconis smilacis sp. nov. Figs. 66-68

Rupturing the periderm as numerous circular conic pustules 1-2 mm in diameter with a central gray disc, 0.5-1 mm in diameter, through which the short stout cylindric ostioles scarcely protrude; perithecia spheric to flattened, 300-500 x 250-350 microns, circinately arranged in the unaltered cortex beneath the well developed truncate-conic grayish ectostroma, and often causing swollen prominences around the central pustule; asci elongate-clavate, with a refractive ring at the apex, 110-125 x 10-16 microns, paraphysate; ascospores uniseriate, broad, ovoid-ellipsoid, with ends somewhat tapered at first but becoming rounded, two-celled, hyaline, becoming tardily brown, constricted at the septum when mature, coarsely granular, 15-21 x 7-8.5 microns.

On Smilax sp., Amana, October 5, 1957, L.H. Tiffany

This species is very close to Melanconis stilbostoma, but differs in that the ostioles scarcely protrude through the grayish disc, the entostroma is grayish without a yellow cast and the tardy browning of the ascospores.

Melanconis smilacis sp. nov.

Stromatibus e basi orbiculari late conoideo-truncatis, pustulatis, salvo disco, peridermio adhaerente; peritheciis in singulo stromate, 2-12 circinantibus, subsphaeroideis; ostiolis elongatis, laevibus, nitidis in disco peridermium cinereo; asci subcylindracei vel elongatis-clavatis,

110-125 x 10-16 microns; ascosporae octonis, monostichis, ellipsoideis, uniseptatis, medio constrictis, hyalinis, tandem olivaceis, 15-21 x 7-8.5 microns.

Hab. in ramis emortuis Smilacis sp.

12. Melanconis appendiculata (Otth) Wehm. Figs. 231-234

On the surface as rather strongly pustulate hemispheric to conic pustules 1-2 mm in diameter, with central rupture exposing a conic or blackened disc through which the cylindric to conic ostioles protrude; perithecia 500-800 x 400-600 microns, circinate in clusters of four to six beneath a slight ectostromatic development which is fused with the ostiolar tips at maturity, occasionally with a blackened marginal zone; asci clavate, 150 x 18-22 microns; ascospores biseriate, ellipsoid to oblong-ellipsoid, two-celled, constricted at the septum, contents granular hyaline at first, becoming brown, 30-38 x 11-14 microns, with rounded ends, often with a small hyaline caplike appendage at each end.

On Acer negundo L., Lake Okoboji, July 21, 1958, L.H. Tiffany

13. Melanconis acrocystis (Pk.) Ell. and Ev. Figs. 247-250

Flattened conic pustules 1-4 x 0.5-2 mm, ostioles short, stout, punctate, black, from one to three erumpent through a lentical, or several erumpent through a small laterally elongate blackened disc; entostroma more or less well developed, conic, gray or gray-green to yellow-green, formed on bark surface just under periderm; perithecia flattened, collapsing, 400-700 x 250-500 microns, crowded circinately beneath the ectostroma and often more or less surrounded by a yellow-brown pseudoparenchymatic entostroma; asci large, clavate, 90-300 x 20-40 microns; spores biseriate to irregularly uniseriate, oblong-ellipsoid, ends rounded, two-celled, slightly constricted at septum, brown, 34-65 x 15-20 microns, with faint globular hyaline caplike appendage at each end; paraphyses bandlike, guttulate, evanescent.

Wehmeyer (1941b) found two conidial types on stromata similar to the perithecial ectostroma; pycnidia with an irregular flattened central cavity containing conidia which were fusoid-ellipsoid, biguttulate, hyaline, one-celled, 5.5-6.5 x 2.5-3.0 microns; locules with conidia cylindric, strongly curved, hyaline, one-celled, 34-42 x 3.5 microns.

On Ulmus americana L., Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany

14. Melanconis decorahensis Ell. Figs. 69-72

Pustules flat, circular-conic, 1-3 mm in diameter, more or less prominent, with central angular to fusoid disc, minute, within a lenticel or 0.2-1.5 x 0.1-0.7 mm, dark gray-green to yellow-green and pulverulent; ostioles stout, black, barely erumpent through disc; perithecia spheric to flattened, 400-800 x 400 microns, circinate within bark; asci long-cylindric, 100-140 x 10-11 microns; ascospores uniseriate to oblique-uniseriate, broad ellipsoid-fusoid, two-celled, brown, constricted at septum, 14-21 x 7-9 microns.

Melanconium conidial stage, conidia ovoid to elongate-ellipsoid or irregular in shape, one-celled, brown, flattened apiculate base, 13-22 x 5-10 microns.

On Betula sp., Decorah, Aug. 11, 1882, E.W. Holway (type); Ellis and Everhart (1892); Wehmeyer (1941b) cites Holway No. 196 on Betula sp. as type.

15. Melanconis corni Wehm. Figs. 243-246

Scarcely visible on surface or visible as minute pustules of papillate erumpent ostioles 0.1-0.2 mm in diameter or as circular perforations; perithecia 400-500 microns in diameter, in small loose groups in unaltered bark cortex, walls thick, parenchymatic; little or no visible entostroma; ostioles convergent and often erumpent as minute disc; asci long-cylindric, with refractive ring in apex, 150-160 x 8-9 μ; paraphyses numerous, broad, bandlike, guttulate; spores overlapping-uniseriate, fusoid-ellipsoid, two-celled, brown, constricted at septum, biguttulate, 16-25 x 7-8.5 microns.

Conidial stage obtained in culture by Wehmeyer (1941b) of loosely compacted stromata formed on surface or within periderm, small spheric or ovoid locules occur with these stromata and contain cylindric slightly tapered or constricted granular hyaline four-celled conidia, 17.5-30 x 7-8 microns, borne on short conidiophores.

On Cornus sp., Fort Defiance State Park, Aug. 8, 1958, L.H. Tiffany

Our collection has ascospores which are only slightly pigmented; however, they are quite young.

16. Melanconis sudans (B. and C.) Wehm. Figs. 73-75

Flat pustulate swellings 0.5-1.5 mm in diameter with minute central disc of fused erumpent ostioles, periderm ruptured to form a perforate or slitlike opening; perithecia 350-550 x 300-450 microns, often lying on sides, parallel to bark surface with short later convergent ostioles; perithecial walls thick, parenchymatous; asci cylindric-clavate, 185-300 x 19-30 microns; paraphyses broad, bandlike, soon evanescent; ascospores biseriate, becoming uniseriate, ellipsoid, two-celled, brown, constricted at septum, finally with large guttula in each cell, 31-52 x 15-21 microns. Asci rupture early, squeezing spores out with envelope of gelatinous periplasm which soon disappears; exuded spores often blackening bark surface.

Melanconiopsis conidial stage; pycnidia formed in entostromata on bark surface; conidia ellipsoid to subglobose with flattened base, one-celled, dark olive-brown, 20-35 x 14-20 microns.

On Acer platanoides L., Ames, 1956, L.H. Tiffany

On Acer saccharinum L., Ames, Dec. 2, 1954, R.M. Lewis; Boone County, May 11, 1958, L.H. Tiffany

On Acer saccharum Marsh., Ames, March 22, 1957, L. H. Tiffany; Ames, Nov. 15, 1957, L.H. Tiffany

On Acer sp., Ledges State Park, Feb. 23, 1958, L.H. Tiffany

On Ulmus americana L., Ames, July 3, 1957, L.H. Tiffany; Amana, Oct. 3, 1957, L.H. Tiffany

8. Diaporthe (Nit.) Wehm.

Perithecia formed within the substratum, erumpent to the exterior through an ectostroma or directly through the overlying tissues by means of a more or less elongated perithecial neck, scattered singly, irregularly clustered or in definitely oriented groups, formed within an area of entostromatic development which shows a marginal blackening of the tissues, at least at some points. Asci clavate to clavate-cylindric, with a refractive ring in the thickened apical wall, sessile, soon freed from their attachment by dissolution of the basal portion and coming to be free within the perithecium. Paraphyses broad, bandlike, present at first, but disappearing with maturity; ascospores fusoid-ellipsoid to cylindric, straight, unequilateral or curved, two-celled, hyaline, sometimes appendaged, and biseriate to uniseriate in the ascus.

Key to the Species of the Genus DIAPORTHE

a. Perithecia scattered. . . . . . . Effusae
  b. On herbaceous stems
    c. Ostiolar necks long-filiform, hairlike; ascospores 8-15 x 2-3.5 microns. . . . 1. D. phaseolorum
    cc. Ostiolar necks short, conic, ascospores 10-17 x 2.5-4 microns. . . . . 2. D. arctii
  bb. On woody stems
    c. Ascospores 1.5-3 microns in diameter. . . . 3. D. prunicola
    cc. Ascospores 2.5 microns or more in diameter
      d. Ascospores 9-15 microns in length
        e. Perithecia scattered
          f. Ostiolar necks short. . . . . 4. D. eres
          ff. Ostiolar necks elongate, filiform. . 5. D. medusaea
        ee. Perithecia in clusters
          f. Ventral zone present. . . . . . . 6. D. beckhausii
          ff. Ventral zone absent
            g. Ascospores fusoid. . . . . . . 7. D. spiculosa
            gg. Ascospores cylindric. . . . . . 8. D. bakeri
      dd. Ascospores 12-18 microns in length
        e. Perithecia scattered
          f. Perithecia 400-720 microns, ascospores 3-4.5 microns. . . . . 9. D. sociabilis var. sambuci
          ff. Perithecia 240-400 microns, ascospores 4-6.5 microns. . . . . 10. D. viburni
        ee. Perithecia clustered, 480-720 microns in diameter
          f. Ostioles not erumpent through a disc. 11. D. decedens
          ff. Ostioles erumpent through a disc. . . 12. D. otthii
aa. Perithecia gathered into groups. Pustulatae
  b. Ectostromata strongly developed as a conic or pulvinate grayish disc
    c. Ascospores less than 3.5 microns in diameter. . . . . 13. D. strumella
    cc. Ascospores more than 3.5 microns in diameter.

d. Ventral zone definitely present in bark or wood
e. Ascospores without appendages. . . . 14. D. leiphaemia var. raveneliana
ee. Ascospores appendaged
f. Entostromata isolate; ascospores with both lateral and apical appendages. . 15. D. taleola
ff. Entostromata effuse, ascospores with apical appendages. . . . . 16. D. pruni
dd. Ventral zone not present. . . . . 14. D. leiphaemia var. raveneliana
bb. Ectostromata poorly developed
c. Ascospores not more than 5.5 microns in diameter
d. Entostromata limited in area. . . . . 17. D. acerina
dd. Entostromata not limited in area
e. Ascospores 10-15 microns in length
f. Ascospores not over 4 microns in diameter
g. Ventral zones present. . . . . 6. D. beckhausii
gg. Ventral zones absent. . . . . . 7. D. spiculosa
ff. Ascospores 4-5 microns in diameter. 17. D. acerina
ee. Ascospores more than 15 microns in length
f. Ascospores 13-25 microns in length
g. Ascospores somewhat inequilateral or curved. . . . . 18. D. melanocarpa
gg. Ascospores straight
h. On Robinia. . . . . 19. D. oncostoma
hh. On Prunus. . . . . 20. D. padi
ff. Ascospores over 25 microns in length. 21. D. peckii
cc. Ascospores more than 5.5 microns in diameter
d. Ascospores appendaged
e. Appendages lateral and apical. . . . . . 15. D. taleola
ee. Appendages apical only. . . . . 22. D. tessella
dd. Ascospores not appendaged
e. Ascospores inequilateral or curved. . . 23. D. tiliacea
ee. Ascospores straight
f. Ascospores less than 20 microns in length
g. Ventral zone brownish band. . . 24. D. inaequalis
gg. Ventral zone sharp, black
h. Ventral zones definite and complete. . . . . 25. D. tuberculosa
hh. Ventral zones incomplete. . . . 26. D. dubia
ff. Ascospores more than 20 microns in length
g. Ascospores not over 6 microns in diameter. . . . . 27. D. megalospora
gg. Ascospores over 6 microns in diameter
h. Stromata pustulate-effuse. . . 23. D. tiliacea
hh. Stromata isolate. . . . . . . 15. D. taleola

1. Diaporthe phaseolorum (Cke. and Ell.) Sacc.

Surface more or less blackened over wide areas; ostioles short-conic to elongate-filiform, sinuous, 120-460 x 50-80 microns, erumpent separately; dorsal zone along dark surface, ventral zone usually absent, occasionally present laterally or along pith; perithecia small, 160-350 x 110-200 microns, scattered or crowded; asci clavate, 28-46 x 5.5-5.8 microns; ascospores biseriate, broad-fusoid, two-celled, hyaline, constricted at septum, 8-12 x 2-3.5 microns.

Phomopsis pycnidial stage; alpha conidia oblong to fusoid, one-celled, hyaline, 5.1-8.5 x 1.7-4 microns; beta conidia long-cylindric, straight or curved, one-celled, hyaline, 11-31 x 1.3-2.4 microns.

On Phalaris arundinacea L., Lake Okoboji, Aug., 1956, L.H. Tiffany

Diaporthe phaseolorum var. sojae (Lehm.) Wehm. Figs. 97-100

Perithecia spherical or mutually compressed laterally, immersed in black stromata, 145-348 x 116-318 microns; beak very long, slender, slightly tapering, 40-60 x 1.5 microns, black; wall definite, outer layer black, inner layer hyaline; asci sessile, elongate, clavate, 37-50 x 7-12 microns; ascospores hyaline, elongate-ellipsoid, two-celled, slightly or not at all constricted at septum, possessing 2-4 guttulae, 10-18.5 x 3.5-5.5 microns. Heterothallic.

Phomopsis pycnidial stage connected in culture by Lehman (1932); pycnidia lenticular, subglobose, subepidermal, simple or chambered, 82-225 x 82-375 microns, beak short; alpha conidia oblong to fusiform, one-celled, hyaline, 6-7 x 2-2.5 microns; beta conidia seldom present, slender, curved, or hooked.

On Glycine max (L.) Merr., Ames, March 1, 1947, A.W. Welch

The ascospore measurements of our specimens as reported by Welch and Gilman (1948) are somewhat larger than those given by Wehmeyer.

Diaporthe phaseolorum var. batatatis (Harter and Field) Wehm. var. caulivora Athow and Caldwell (1954) Figs. 101-104

Perithecia grouped in valsoid manner, stromata immersed erumpent; perithecia carbonaceous, 9-25 in a stroma, subglobose, immersed, 120-370 microns in diameter; ostioles erumpent, elongate, tapering, 1-3 mm; asci clavate-cylindric, sessile, 27-40 x 6.5-8.5 microns; ascospores biseriate to obliquely uniseriate, subellipsoid, rounded at ends, obtuse, obtuse, constricted at septum, 8.5-10.5 x 3.5-5 microns. Homothallic.

Phomopsis pycnidial stage; pycnidia irregularly chambered, erumpent, 60-130 x 60-110 microns; alpha conidia oblong-fusoid, 6-8 x 3-5 microns; beta conidia filiform-hamate, 16-30 x 1.3-2.4 microns.

On Glycine max (L.) Merr., Ames, Feb. 10, 1947, A.W. Welch

2. Diaporthe arctii (Lasch) Nit. Figs. 105-108

Surface slightly, irregularly or heavily blackened areas widely effuse or confluent; dorsal blackened zones usually developed on bark surface but often masked by overlying epidermis; ostioles cylindric, erumpent singly or in small loose groups; ventral zone present at least at margin of fruiting areas; perithecia spheric to somewhat flattened, 280-480 x 160-320 microns, usually buried; asci clavate, refractive ring in apex,

47-60 x 7-10 microns; ascospores biseriate, fusoid-ellipsoid, straight or more or less inequilateral or curved, two-celled, hyaline, constricted at septum when mature, 12-15 x 2.5-4 microns.

Phomopsis pycnidial stage; alpha conidia, 7-10 x 2-3 microns; beta conidia 18-30 x 1.0-1.5 microns.

On Ambrosia trifida L., Ames, Oct. 5, 1955, R.M. Lewis; Ledges State Park, July 25, 1957, L.H. Tiffany
On Arctium sp., Amana, Oct. 3, 1957, L.H. Tiffany
On Asclepsias syriaca L., Decorah, July, 1884, E.W. Holway; Ellis and Everhart (1892) as Diaporthe asclepiadis Ell. and Ev.
On dead stem of Chenopodium sp., Ames, March, 1925, J.C. Gilman (Diaporthe euspina (C. and E.) Sacc.)
On dead stem of Erigeron sp., Decorah, July, 1882, E.W. Holway
On some large herbaceous stem, E.W. Holway (Diaporthe eburensis Sacc.) Ellis and Everhart (1892)
On stems of some composite plant, Decorah, July, 1892, E.W. Holway
On overwintered herbaceous stem, Ames, Sept. 17, 1957, L.H. Tiffany; Ames, Oct. 25, 1957, L.H. Tiffany; Fort Defiance State Park, Aug. 20, 1958, L.H. Tiffany; Lake Okoboji Lakeside Lab., July 23, 1958, L.H. Tiffany; Ledges State Park, Sept. 8, 1957, L.H. Tiffany; Spillville, June 1, 1958, L.H. Tiffany

3. Diaporthe prunicola (Pk.) Wehm. Figs. 109-112

On surface as circular or laterally elongate, pustulate ruptures of the periderm through which are erumpent compact discs composed of fascicles of stout-cylindric, punctate ostioles, surface of bark characteristically blackened and covered with numerous small, black, carbonaceous papillae; ventral zone in wood; perithecia 400-720 x 320-600 microns, in definite clusters within pustulate swollen areas of entostroma; asci clavate, 40-55 x 6-8 microns; ascospores biseriate to triseriate, oblong-cylindric with rounded ends, hyaline, becoming two-celled at full maturity, straight or slightly curved, very slightly if at all constricted, four guttulate, 11-13 x 1.5-2 microns.

On Prunus americana Marsh., Decorah, May, 1888, E.W. Holway (Diaporthe cylindrospora Pk.); Ellis and Everhart (1892) as Cryptospora pennsylvanica
On Prunus serotina Ehrh., Collins, Sept. 21, 1958, L.H. Tiffany
On dead limbs of Prunus sp., Decorah, May 1, 1892, E.W. Holway (Diaporthe pruni Ell. and Ev.)

4. Diaporthe eres Nit. Figs. 113-116

On surface as small pustulate ruptures or angular perforations of the periderm, often exposing the blackened surface of the bark; ostioles short cylindric, erumpent singly or in small loose clusters; ventral zones always present at margins of fruiting areas and usually more or less complete beneath; perithecia spherical or flattened, 240-800 x 160-500 microns, scattered singly, buried in either bark or wood; asci clavate, refractive ring in apex, 40-60 x 5-8 microns; ascospores biseriate, hyaline, long, narrow-fusoid to fusoid, often inequilateral, two-celled, constricted at septum, 9.5-15 x 2.5-4 microns.

Phomopsis pycnidial stage; alpha conidia oblong fusoid, 7-10 x 2-3 microns; beta conidia filiform, curved, 20-27 x 1 micron.

On Acer sp. (soft maple), Iowa City, Oct. 3, 1924, G. W. Martin (SUI)

On Cornus paniculata l'Her, Decorah, June 9, 1892, E.W. Holway (Diaporthe cornicola Ell. and Holw.); Wehmeyer (1933)

On Crataegus crus-galli L., Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany; Marble Lake, Aug. 1, 1958, L.H. Tiffany

On Fraxinus pennsylvanica var. lanceolata (Borkh.) Sarg., Ledges State Park, July 23, 1957, L.H. Tiffany

On Lonicera tatarica L., Ames, Oct. 13, 1957, L.H. Tiffany; Lake Okoboji Lakeside Lab., July 28, 1958, L.H. Tiffany

On Physocarpus opulifolius (L.) Maxim, Cedar Falls, J.A. Parrish (SUI)

On Prunus sp., Milford Woods, July 22, 1958, L.H. Tiffany

On Pyrus sp., Ames, Dec. 1, 1954, R.M. Lewis

On Quercus sp., Ames, Jan. 30, 1958, L.H. Tiffany; Estherville, July 18, 1956, L.H. Tiffany; Johnson County, 1907, J. A. Parrish (SUI)

On Rhamnus cathartica L., Ames, July 8, 1958, L.H. Tiffany

On Tilia americana L., Ledges State Park, Feb. 23, 1958, L.H. Tiffany

On Ulmus americana L., Call State Park, Aug. 13, 1958, L.H. Tiffany; Milford Woods, Aug. 15, 1958, L.H. Tiffany

On Ulmus sp., Ames, Aug., 1954, R.M. Lewis; Fort Dodge, June 16, 1956, L.H. Tiffany; Lake Okoboji, Aug. 1, 1956, L.H. Tiffany

5. Diaporthe medusaea Nit. Figs. 117-120

On surface as elongate-cylindric or sinuous-filiform ostioles, often appressed against or beneath the periderm, erumpent singly or loosely aggregated in large clusters, not united in a definite disc, bark surface in some forms heavily blackened, in others not; perithecia 200-500 microns in diameter, irregularly scattered or tending to be in groups or crowded clusters, often forming somewhat pustulate swellings; asci clavate with refractive ring in apex, 40-47 x 6-9 microns; ascospores biseriate, hyaline, two-celled, fusoid-ellipsoid, 10-15 x 2.5-3.5 microns.

Phomopsis pycnidial stage; alpha conidia ovoid-fusoid to elongate, 5-9 x 2-3 microns; beta conidia slender, 20-36 x 1.0-1.5 microns.

On Caragana sp., Ames, June 14, 1955, R.M. Lewis

On Fraxinus pennsylvanica var. lanceolata (Borkh.) Sarg., Woodman Hollow State Park, Oct. 5, 1958, L.H. Tiffany

On Juglans regia L. (?), Cedar Falls, April 27, 1957, W.H. Bragonier

On Ulmus americana L., Dolliver State Park, Oct. 5, 1958, L.H. Tiffany

On Ulmus sp., Ames, July 8, 1958, L.H. Tiffany; Ledges State Park, June, 1957, L.H. Tiffany

On Vitis labrusca L., Dolliver State Park, Oct. 5, 1958, L.H. Tiffany

On Xanthoxylum americanum L., Pilot Knob State Park, April 16, 1957, L.H. Tiffany

The Ames collection on Ulmus has slightly larger ascospores, but agrees well in other details with D. medusaea, It compares favorably with Ellis and Everhart N.A.F. 2533.

6. Diaporthe beckhausii Nit. Figs. 121-124

On surface as small pustulate ruptures with one or a few barely erumpent cylindric ostioles or as small definite circular to fusoid often laterally elongated discs of short-cylindric ostioles; dorsal zone often absent when perithecia thickly crowded but usually present and irregularly pustulate, dipping irregularly into bark with tendency to run within the wood; perithecia 250-720 x 250-500 microns, scattered singly, irregularly crowded, or loosely grouped, usually collectively erumpent; asci clavate, 40-48 x 5-7 microns; ascospores biseriate, fusoid-ellipsoid, straight, constricted at septum, 10-13 x 2.5-3.5 microns.

Phomopsis pycnidial stage; alpha conidia fusoid-ellipsoid, one-celled, hyaline, two-guttulate, 8.5-12 and 2-3 microns; beta conidia fusoid-cylindric, crescent-shaped, one-celled, hyaline, 8-13 x 1.0-1.5 microns.

On Betula alleghaniensis Britton, Ames, April 10, 1958, L.H. Tiffany

On Betula nigra L., Ames, Oct. 12, 1955, R.M. Lewis

On Fraxinus pennsylvanica var. lanceolata (Borkh.) Sarg., Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany

On Betula, the dorsal zone is absent or pustulate with the perithecia tending to be in small clusters and collectively erumpent.

7. Diaporthe spiculosa (Alb. and Schw.) Nit. Figs. 125-128

On surface as compact circular pustulate discs of cylindric ostioles, stromata widely effuse; dorsal zone often faint or definitely absent; no ventral zone present; perithecia 200-500 microns in diameter, definitely clustered in small groups and erumpent collectively; asci clavate with refractive ring in apex, 40-47 x 6-9 microns; ascospores biseriate, fusoid-ellipsoid, two-celled, hyaline, 12-15 x 2.5-4 microns.

Phomopsis pycnidia with conidia oblong-ovoid to oblong-fusoid, 8-10 x 3-4 microns.

On Juglans cinerea L., Decorah, July 2, 1882, E.W. Holway

On dead limbs of Juglans cinerea L., Decorah, June, 1892, E.W. Holway; (Ell. and Ev. N.A.F. 2817 as Diaporthe bicincta (C. and P.) Sacc.):Wehmeyer (1933)

On Juglans nigra L., Amana, Oct. 3, 1957, L.H. Tiffany

On Juglans sp., Ledges State Park, July 1, 1957, L.H. Tiffany

On Quercus alba L., Boone, Oct. 13, 1957, L.H. Tiffany

On Tilia americana L., Call State Park, Aug. 13, 1958, D. Humphreys

8. Diaporthe bakeri Wehm. Figs. 129-132

On surface as numerous dense irregular clusters of short-cylindric ostioles, often longitudinally confluent and erumpent through irregular pustulate ruptures of the periderm; entostromata irregularly pustulate-effuse, differentiated, lighter in color; dorsal zone present along bark surface when pustulate areas are confluent or crowded but dipping into bark or wood where more widely spaced, no ventral zone; perithecia spherical, 400-480 microns in diameter, usually grouped in large loose

clusters in pustulate entostromatic areas which may be confluent and appear effuse; asci clavate, 40-46 x 5-8 microns; spores biseriate, cylindric to oblong-fusoid, rounded at ends, two-celled, hyaline, not constructed at septum, 9-13 x 2-3 microns.

On Carpinus caroliniana Walt., Ledges State Park, July, 1957, L.H. Tiffany

9. Diaporthe sociabilis Nit. var. sambuci (Ell. and Ev.) Wehm.
Figs. 227-230

Ruptures periderm by formation of numerous small blackened ectostromata which are often more or less seriately arranged; surface of bark more or less blackened, dorsal zone present, blackening not pustulate; ostioles barely erumpent; no ventral zone present; perithecia 400-720 x 300-400 microns, irregularly scattered or definitely grouped, usually collectively erumpent; asci clavate, refractive ring in apex, 60-80 x 8-9 microns; ascospores biseriate, fusoid-ellipsoid, two-celled, hyaline, constricted at septum, 14-17.5 x 2.8-4µ, young spores may have short hyaline, evanescent appendages and slight gelatinous envelope.

On Sambucis canadensis L., Ames, July 1, 1958, L.H. Tiffany
Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany

This species is very like Diaporthe spiculosa but differs from that species by having a definite blackened dorsal zone, larger perithecia and ascospores.

10. Diaporthe viburni Dearn. and Bisby Figs. 133-136

On surface as numerous crowded, papillate pustules, 0.1-0.2 mm in diameter, short ostioles barely erumpent singly or in small groups; dorsal blackening absent or present merely as blackening above perithecia, or as short patches of dorsal zone dipping into bark; no ventral zone; perithecia 240-320 microns in diameter, thickly but irregularly scattered; asci clavate with refractive ring in apex, 55-67 x 8-12 microns; ascospores biseriate, fusoid-ellipsoid to oblong-ellipsoid, two-celled, hyaline, slightly constricted at septum at maturity, 14-18 x 4-6 microns.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933); alpha conidia fusoid-ellipsoid, one-celled, hyaline, 8-12 x 2-3 microns; beta conidia long-fusoid, hamate, one-celled, hyaline, 15-33 x 1 micron.

On dead limbs of Viburnum lentago L., Decorah, June, 1882, E.W. Holway (as Diaporthe beckhausii Nit.); Wehmeyer (1933)
On Viburnum sp., Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany

11. Diaporthe decedens (Fr.) Fckl. Figs. 137-140

On surface as numerous small circular pustulate discs 0.2-0.4 mm in diameter through which papillate ostioles become separately erumpent forming scattered groups; no blackened zone in either wood or bark; perithecia spherical or flattened, 480-720 x 320-480 microns, in loose clusters but always separately erumpent; asci clavate with refractive ring at apex, 65-85 x 10-15 microns; ascospores fusoid-ellipsoid, variable in size, 13-22 x 3.5-6 microns, faint hyaline appendages sometimes present on spores.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933);

alpha conidia fusoid-ellipsoid to elongate-fusoid, straight, one-celled, hyaline, 11-15 x 2-2.5 microns; beta conidia narrow-cylindric, hyaline, one-celled, curved or allantoid, 8-13 x 1-1.5 microns.

On Celtis occidentalis L., Ames, Dec., 1954, R.M. Lewis

On Corylus americana Walt., Fort Defiance State Park, July 30, 1958, L.H. Tiffany; Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany; Ledges State Park, June 4, 1958, L.H. Tiffany; Milford Woods, Aug. 15, 1958, L.H. Tiffany

On dead limbs of Corylus, Iowa, E.W. Holway; Ellis and Everhart (1892) as Diaporthe tessera (Fr.) Fckl.; Wehmeyer (1933)

On Corylus sp., Decorah, June 7, 1892, E.W. Holway (Ell. and Ev. N.A.F. 2818); Fort Defiance State Park, July 22, 1956, L.H. Tiffany

On unknown host, Iowa, MacBride (SUI)

12. Diaporthe otthii Nit. Figs. 235-238

Conic pustulate stromata 0.5-1 mm in diameter with central circular disc of black cylindric ostioles; slight conic ectostroma, bark surface usually strongly blackened at point of emergence of ostioles; entostromata widely effuse, strongly differentiated; dorsal zone occasionally present along bark surface, when present usually dipping slightly into bark; ventral zone definitely present well within wood; perithecia 400-720 x 400-480 microns, definitely clustered, often pustulate areas, collectively erumpent; asci clavate, 70-80 x 10-15 microns, ascospores biseriate, fusoid-ellipsoid, two-celled, hyaline, four-guttulate, often strongly constricted at septum, 16-25 x 4-8 microns.

On Ulmus americana L., Fort Defiance State Park, Aug. 8, 1958, L.H. Tiffany; Lake Okoboji Lakeside Lab., July 28, 1958, L.H. Tiffany

13. Diaporthe strumella (Fr.) Fckl. Figs. 141-144

On surface as scattered, pustulate, circular or laterally elongated, blackened, stromatic discs, 0.5-2 x 0.2-1 mm containing scattered papillate to conic ostioles, or discs nearly obliterated by dense fascicle of ostioles which are occasionally much elongated and bent; perithecia spherical to ovoid, 250-400 microns in diameter, definitely clustered; usually no darkened zones visible in either bark or wood, obscure dorsal zone which runs between bark and wood, may be blackened along margin of pith; ventral zones sometimes deep in wood; asci clavate, with refractive ring in apex, 37-45 x 6-9 microns; ascospores biseriate, fusoid, often somewhat inequilateral or curved, two-celled, hyaline, slightly constricted at septum at maturity, 11-15 x 2-3 microns.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933); alpha conidia fusoid-ellipsoid, one-celled, hyaline, 6-8 x 2.5 microns; beta conidia long fusoid-cylindric, straight, or slightly curved, one-celled, hyaline, 11-15 x 1.5 microns.

On Ribes sp., Lake Okoboji Lakeside Lab., Aug. 1, 1956, L.H. Tiffany; Lake Okoboji Lakeside Lab., July 23, 1958, L.H. Tiffany

On Ribes missouriense Nutt., Fort Defiance State Park, Aug. 20, 1958, L.H. Tiffany; Lake Okoboji Lakeside Lab., July 28, 1958, L.H. Tiffany

14. Diaporthe leiphaemia var. raveneliana (DeThuem. and Rehm) Wehm. Figs. 145-148

On surface as well developed pulvinate stromata which rupture the periderm in angular fashion exposing erumpent circular to angular orange-yellow to brownish discs, discs becoming cracked and roughened in age; ostioles scarcely erumpent; no blackened dorsal zone; perithecia radially elongated, 240-480 x 320-640 microns, clustered beneath ectostromatic discs, surrounded by rich development of ectostromatic mycelium; asci clavate with refractive ring in apex, 55-65 x 6-9 microns; ascospores biseriate, fusoid, ellipsoid, hyaline, two-celled, slightly constricted at septum, 12-15 x 4-6 microns.

Phomopsis pycnidia obtained in culture by Wehmeyer (1933); alpha conidia long, fusoid-cylindric, one-celled, hyaline, 11-20 x 2-5 microns; beta conidia short cylindric to allantoid, one-celled, hyaline, 5.5-10 x 1.5-2 microns.

On Quercus sp., Decorah, May, 1883, E.W. Holway, Ellis and Everhart (1892) as Diaporthe taleola (Fr.) Sacc.; Woodman Hollow State Park, Oct. 5, 1958, L.H. Tiffany

On Quercus alba L., Amana, Oct. 3, 1957, L.H. Tiffany; Ames, Feb. 28, 1955, R.M. Lewis; Ames, Oct. 25, 1957, L.H. Tiffany; Fort Defiance State Park, July 30, 1958, L.H. Tiffany; Fort Defiance State Park, Aug. 8, 1958, L.H. Tiffany; Ledges State Park, July 23, 1957, L.H. Tiffany; Pilot Knob State Park, July 25, 1956, L.H. Tiffany; Pine Lake State Park, Sept. 15, 1957, L.H. Tiffany

Some confusion has arisen concerning the species of Diaporthe on Quercus. Both Diaporthe leiphaemia var. raveneliana and Diaporthe taleola have been found widespread in the state.

15. Diaporthe taleola (Fr.) Sacc. Figs. 155-158

Appearing on surface as small pustules with closely adherent periderm and central circular to fusoid whitish or blackened disc, 0.2-1 mm in diameter; stout hemispheric clustered ostioles sometimes erumpent; no dorsal zone present; sharp ventral zone entirely within the bark, sometimes abuts upon periderm appearing upon surface as marginal ridge about pustule; perithecia mostly radially elongated, 240-400 x 320-400 microns, arranged in definite cluster within isolate entostromatic areas and beneath a definite white conic to pulvinate ectostroma; asci long cylindric with stipelike base and refractive ring in apex, usually eight but sometimes four-spored, 130-160 x 10-13 microns; ascospores uniseriate, broad-ellipsoid, two-celled, hyaline, 17-25 x 7-9 microns, constricted at septum, with cylindric hyaline, apical appendages, 6-10 x 1-1.5 microns and 2-3 lateral, somewhat longer appendages radiating from region of septum.

Conidial stage probably a Myxosporium with conidia cylindric, curved, one-celled, hyaline, 20-30 x 4-5 microns.

On Quercus alba L., Ames, Nov. 15, 1957, L.H. Tiffany; Fort Defiance State Park, Aug. 20, 1958, L.H. Tiffany

On Quercus macrocarpa Michx., Ames, Sept. 5, 1955, R.M. Lewis; Decorah, May 9, 1886, E.W. Holway; Gull Point State Park, July 24, 1958, L.H. Tiffany; Fort Defiance State Park, July 30, 1958, L.H. Tiffany

On Quercus sp., Amana, Oct. 3, 1957, L.H. Tiffany; Ames, April 9, 1955, R.M. Lewis; Ames, March 19, 1956, L.H. Tiffany; Ames, March 23, 1958, L.H. Tiffany; Ledges State Park, July 10, 1957, L.H. Tiffany; Ledges State Park, March 4, 1958, L.H. Tiffany

On dead oak limbs, Iowa, E.W. Holway in Ellis and Everhart (1892)

Wehmeyer (1933) considered Diaporthe taleola as an European species and transferred the American specimens to D. leiphaemia var. raveneliana. Our Iowa specimens correspond very closely with the descriptions of D. taleola and all have the characteristic appendages on the ascospores (Gilman, Tiffany, and Lichtwardt (1957)).

16. Diaporthe pruni Ell. and Ev. Figs. 149-154

On surface as pustulate, conic, mostly laterally elongate stromata with central circular to elliptic, whitish to blackened disc, 1-1.5 x 0.3-0.7 mm in diameter through which cluster of cylindric ostioles is erumpent; ventral zone definite within wood; perithecia 500-700 x 300-500 microns, usually partly or entirely sunken in wood; asci clavate, 65-92 x 6-10 microns; ascospores biseriate, fusoid-ellipsoid, two-celled, hyaline, constricted at septum, 15-20 x 4.5-5 microns with short thick, hyaline, evanescent appendage 4-8 microns long at each end.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933); alpha conidia fusoid-ellipsoid, one-celled, hyaline, 10-16 x 2.5-3 μ; beta conidia long-cylindric, hamate, one-celled, hyaline, 10-15 x 1-1.5 μ.

On Prunus serotina Ehrh., Decorah, May 1, 1892, E.W. Holway

On Prunus virginiana L., Gull Point State Park, July 24, 1958, L.H Tiffany

The species of Diaporthe on Prunus have been much confused. Of the three collections cited by Gilman and Archer (1929) under Diaporthe pruni Ell. and Ev. only one remains under this name. The other two are referred to D. padi and D. prunicola, respectively.

Cfr. Ell. and Ev. N.A.F. 2822.

17. Diaporthe acerina (Pk.) Sacc.

On surface as numerous pustulate ruptures of periderm, stout-cylindric ostioles barely erumpent in small loose clusters; dorsal zone definite, dipping into bark between perithecial groups; ventral zone definite and complete within wood; perithecia 300-500 x 225-300 microns, irregularly scattered or in loose groups within pustulate areas but usually collectively erumpent; asci clavate with refractive ring in apex, 60-72 x 7-10 microns; ascospores biseriate, fusoid-ellipsoid, two-celled, hyaline, not constricted except at full maturity, 12-15 x 3-5 microns.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933); alpha conidia fusoid-ellipsoid, one-celled, hyaline, 8-12 x 2-3.5 microns; beta conidia long-cylindric, one-celled, hyaline, straight or curved, 16-27 x 1.0-1.5 microns.

On Acer saccharinum L., Grand River, Sept., 1933, Stark; Ledges State Park, July 10, 1958, L.H. Tiffany

Wehmeyer (1933) states that this species is confined to Acer spicatum Lam. Our material is most closely like the descriptions of this species.

18. Diaporthe melanocarpa Dearn. Figs. 239-242

Small conic pustules with a central minute disc, 0.2-0.4 mm in diameter, consisting of compact cluster of few short-cylindric ostioles; entostromata pustulate-effuse, strongly differentiated, pustulate areas rather widely scattered; dorsal zone definite, dipping to wood surface; ventral zone definite within wood; perithecia 400-520 x 320-400 microns, clustered in small groups within pustulate areas; asci clavate with refractive ring in apex, 60-70 x 6-8 microns; spores biseriate, fusoid-ellipsoid, two-celled, hyaline, usually somewhat inequilateral or curved, slightly constricted at septum, 16-23 x 2.5-4.5 microns.

On Amelanchier canadensis (L.) Medic., Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany

19. Diaporthe oncostoma (Duby) Fckl. Figs. 163-166

On surface as more or less pustulate ruptures of periderm through which stout, short cylindric ostioles are erumpent in small loose clusters or somewhat larger irregular groups; pycnidial stromata often present, forming papillate pustules which later open to exterior; entostromata strongly differentiated, pustulate effuse to evenly effuse, usually widely extended but sometimes limited; dorsal zone definite, dipping into bark between perithecial clusters; ventral zone usually definite and complete beneath, often interrupted and present only at lateral margins; perithecia 350-600 x 350-550 microns, in more or less definite clusters, occasionally irregularly scattered; asci clavate with refractive ring in apex, 60-80 x 6-9 microns; ascospores biseriate, rather narrow fusoid-ellipsoid, two-celled, hyaline, constricted at septum, 13-17 x 3-4 microns.

Phomopsis pycnidial stage; alpha conidia fusoid-ellipsoid, one-celled, hyaline, 7-13 x 2-3 microns; beta conidia filiform-hamate, hyaline, 13-23 x 1.0-1.5 microns.

On Cladrastis lutea (Michx.) Koch, Ames, Sept. 17, 1957, L.H. Tiffany

On Robinia pseudo-acacia L., Ames, April 4, 1955, R.M. Lewis; Ames, Oct. 17, 1957, L.H. Tiffany; Fort Defiance State Park, Aug. 8, 1958, L.H. Tiffany; Fort Defiance State Park, Aug. 20, 1958, L.H. Tiffany; Ledges State Park, Sept. 8, 1957, L.H. Tiffany; Mill Creek State Park, Aug. 18, 1958, L.H. Tiffany; Pine Lake State Park, Sept. 15, 1957, L.H. Tiffany; Wanata State Park, Aug. 18, 1958, L.H. Tiffany

20. Diaporthe padi Otth Figs. 167-170

On surface as circular to fusoid, pustulate discs 0.2-1 mm in diameter, consisting of compact cluster of short, stout, cylindric, punctate ostioles; dorsal zone dipping to bark between perithecial groups; ventral zones absent, greenish or yellowish discolorations sometimes seen in wood; perithecia usually definitely clustered, sometimes irregularly crowded, 320-640 x 240-400 microns, often collectively erumpent, asci clavate, with refractive ring in apex, 60-75 x 7-10 microns; ascospores biseriate, fusoid-ellipsoid, two-celled, hyaline, constricted at septum, 13-19 x 2.5-4.0 microns.

Conidia produced in pycnidia, fusoid, 9-11 x 3 microns.

On Aesculus hippocastanum L., Ames, July 8, 1957, L.H. Tiffany

On Prunus hortulana Bailey, Ames, Sept., 1924, O.T. Miller (as Diaporthe pruni Ell. and Ev.)
On Prunus virginiana L., Lake Okoboji Lakeside Lab., July 27, 1958, L.H. Tiffany
On Ulmus americana L., Milford Woods, Aug. 15, 1958, L.H. Tiffany

21. Diaporthe peckii Sacc. Figs. 171-174

On surface as irregular, scarcely visible, dirty gray, erumpent discs containing 2-5 papillate ostioles, or as more noticeable perforations or as minute separately erumpent conic ostioles; pycnidia often appear as numerous minute pustulate ruptures of periderm; entostroma effuse; indefinite dorsal blackened zone dips irregularly into bark between perithecia and extends into wood at margins of entostromatic areas; perithecia spherical, 300-600 microns in diameter, scattered or clustered in small groups; asci clavate, 65-85 x 8-10 microns; ascospores biseriate fusoid-ellipsoid to fusoid-cylindric, usually somewhat curved and often irregular in shape, two-celled, hyaline, constricted at septum, 20-40 x 3-5 microns.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933); alpha conidia fusoid-ellipsoid, one-celled, hyaline, 9-12 x 2.0-2.5 microns; beta conidia filiform-hamate, one-celled, hyaline, 16-23 x 1.5 microns.

On Rhus sp., Ledges State Park, March 3, 1957, L.H. Tiffany

22. Diaporthe tessella (Pers.) Rehm Figs. 175-177

On surface as clusters of separately erumpent, black, papillate ostioles which are usually grouped about a minute perforation of periderm; sharp black zone extends from margins of area into bark tissue; no ventral zone within wood; perithecia irregularly spherical, 400-725 microns in diameter, occur singly or in groups of 2-8 within pustulate stromatic areas, 1-4 mm in diameter, separately erumpent through periderm; asci clavate with refractive ring in apex, 110-115 x 18-21 microns; ascospores fusoid-cylindric, usually curved or bent at septum, two-celled, hyaline, constricted at septum, 35-55 x 7-9 microns, often possess faint short, hyaline, appendage at each end.

Stromatic pycnidia obtained in culture by Wehmeyer (1933); conidia inequilateral to curved-fusoid or allantoid, one-celled, hyaline, 5.5-9 x 1.0-1.5 microns.

On Salix nigra Marsh., Lake Okoboji Lakeside Lab., Aug., 1958, L.H. Tiffany
On Salix sp., Decorah, May 30, 1892, E.W. Holway; Ellis and Everhart (1892)

23. Diaporthe tiliacea (Ell.) Rehm Figs. 178-180

On surface as numerous minute conic, pustulate ruptures of periderm exposing disclike cluster of short or somewhat elongated cylindric black ostioles 0.1-0.6 mm in diameter; perithecial clusters usually bounded by a more or less definite dorsal and lateral blackened zone which is irregularly pustulate about the confluent stromata; usually broad brownish discolored ventral zone within wood; perithecia spherical or radially

elongated, 320-600 x 320-560 microns, grouped in clusters of 3-15; asci clavate with refractive ring in apex, 90-120 x 13-20 microns; ascospores biseriate, fusoid-ellipsoid, straight or slightly curved, constricted at septum, 24-39 x 6-9.5 microns.

Fusicoccum or Septomyxa type of fruiting body obtained in culture by Wehmeyer (1933); conidia long-fusoid to cylindric, granular, hyaline, 31-45 x 4-5 microns, becoming one- to two-septate.

On Tilia americana L., Decorah, May, 1892, E.W. Holway (Ell. and Ev. N.A.F. 2826 as Melanconis tiliacea Ell. (Ell. and Ev.), Wehmeyer (1933); Fort Defiance State Park, Aug. 8, 1958, L.H. Tiffany; Fort Defiance State Park, Aug. 20, 1958, L.H. Tiffany; Iowa, T.H. Macbride (SUI); Lake Okoboji Lakeside Lab., Aug. 5, 1956, L.H. Tiffany; Lake Okoboji Lakeside Lab., July 25, 1958; L.H. Tiffany; Ledges State Park, March 3, 1957, L.H. Tiffany

On dead Tilia americana L., Iowa, J.C. Arthur:Ellis and Everhart (1892)

This species was described by Ellis (1883a) from Iowa material collected at Ames by J.C. Arthur, Oct., 1882 as Diatrype tiliacea; later Ellis and Everhart transferred it to the genus Melanconis.

24. Diaporthe inaequalis (Curr.) Nit. Figs. 216-219

Small pustulate stromata 0.2-0.5 mm in diameter, erumpent as small clusters of short stout cylindric or elongate, bent ostioles; entostromata pustulate-effuse, dorsal zone running along or just beneath the bark surface and dipping into the bark or wood between perithecial groups; perithecia flattened-spherical, 450-720 x 240-500 microns, scattered singly or clustered, usually collectively erumpent; ventral zone absent or present as a rather broad brownish zone well within the wood; asci clavate, becoming cylindric, 70-110 x 9-15 microns; ascospores biseriate, becoming uniseriate, broadly ellipsoid, hyaline, two-celled, constricted at the septum, cells each with a single large globule, 13-17 x 5-9 microns.

Wehmeyer (1933) includes as conidial forms a Phomopsis with filiform, curved conidia, 21-27 x 2 microns, and a Phoma with elongate-ellipsoid conidia which are inequilateral and 7-10 x 2-3 microns.

On Amorpha fruticosa L., Ames, May 28, 1958, L.H. Tiffany; Marble Lake, Aug. 11, 1958, L.H. Tiffany

On Amorpha sp., Ames, April 12, 1958, L.H. Tiffany

25. Diaporthe tuberculosa (Ell.) Sacc. Figs. 181-184

On surface as small loose clusters of slightly erumpent ostioles, often producing small circular perforations of periderm; entostromata pustulate-effuse or occasionally isolate; ventral zone continuous beneath, usually well within wood; perithecia spherical, 400-600 microns in diameter, in small groups within the pustulate areas or sometimes irregularly gregarious in the more evenly effuse areas, but nearly always collectively erumpent; asci clavate with refractive ring in apex, 60-80 x 10-14 microns; ascospores biseriate, fusoid-ellipsoid, obtuse, two-celled, hyaline, constricted at septum, 14-17 x 4.5-7.5 microns.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933); alpha conidia fusoid-ellipsoid, one-celled, hyaline, 7-12 x 3.5-4.5 mi-

crons; beta conidia crescent-shaped with tapered ends, 11-20 x 1.3-2 microns.

On Amelanchier canadensis (L.) Medic., Ames, June 23, 1957, L.H. Tiffany; Ames, Oct. 25, 1957, L.H. Tiffany; Boone County, June 9, 1958, L.H. Tiffany; Dolliver State Park, Oct. 5, 1958, L.H. Tiffany; Ledges State Park, Nov. 30, 1957, L.H. Tiffany

D. tuberculosa is similar to D. beckhausii, but has larger spores.

26. Diaporthe dubia Nit. Figs. 185-188

On surface as hemispheric to flattened conic pustules through which are erumpent small or irregular clusters of short, stout ostioles; dorsal zone when present dipping into bark between perithecia; ventral zone sometimes complete particularly in the isolate stromata, usually incomplete, present only laterally or entirely lacking; perithecia 300-600 microns in diameter, definitely clustered in pustulate areas, collectively erumpent; asci clavate with refractive ring in apex, 65-85 x 9-15 microns; ascospores biseriate, broad fusoid-ellipsoid, two-celled, hyaline, somewhat constricted at septum at maturity, 13-16 x 4-7 microns.

Phomopsis pycnidial stage obtained in culture by Wehmeyer (1933); alpha conidia fusoid-ellipsoid, one-celled, hyaline, 9.5-13 x 2.5-4 microns; beta conidia short-cylindric, straight or slightly curved, one-celled, hyaline, 9-12 x 1.3-1.5 microns.

On Acer saccharum Marsh., Ames, Sept., 1955, L.H. Tiffany; Ames, May 21, 1957, L.H. Tiffany; Fort Defiance State Park, Aug. 8, 1958, L.H. Tiffany; Woodman Hollow State Park, Oct. 5, 1958, L.H. Tiffany

27. Diaporthe megalospora Ell. and Ev. Figs. 220-222

Often scarcely visible on the surface as scattered slightly erumpent ostioles, short conic to cylindric, occasionally elongate beaks; entostromata irregularly pustulate-effuse; dorsal zone present in bark or along wood often penetrating to form pustulate areas; ventral zones absent or confined to the lateral margins, or as indefinite greenish-black areas in the wood; asci clavate with a refractive ring in the apex, 65-90 x 10-15 microns; ascospores biseriate, fusoid cylindric, two-celled, hyaline, usually slightly curved or inequilateral, constricted at the septum, 24-39 x 3.5-6 microns.

Phomopsis conidial stage obtained in culture by Wehmeyer (1933); alpha conidial ellipsoid to fusoid, one-celled, hyaline, 10-15 x 3-6 microns; beta conidia long-cylindric, hamate, one-celled, hyaline, 14-20 x 1 micron.

On Sambucus canadensis L., Ames, June 8, 1958, L.H. Tiffany; Fort Defiance State Park, Aug. 4, 1958, L.H. Tiffany; Lake Okoboji Lakeside Lab., July 25, 1958, L.H. Tiffany; Wanata State Park, Aug. 18, 1958, L.H. Tiffany

9. Cryptodiaporthe (Petrak) Wehm.

Perithecia immersed in the bark, more or less irregularly scattered or in definite clusters, but usually with convergent ostiolar necks which are erumpent through the periderm or through variously formed ecto-

stromata. Ectostromata scantily developed or as definite conic to pulvinate erumpent discs, or as a loose weft of hyphae causing broad angular ruptures of the periderm. Entostromata very scanty or as a rich development of hyphae about the perithecia, often forming definitely oriented stromata. No blackened marginal zones within the substratum. Asci clavate, often with a refractive ring in the apex and with a tapering base which is evanescent. Ascospores hyaline, two-celled, ellipsoid to fusoid, straight or curved and often appendaged. Conidial stages various.

Key to the Species of the Genus CRYPTODIAPORTHE

a. Ascospores appendaged. . . . . 1. C. aculeans
aa. Ascospores not appendaged
  b. Ascospores inequilateral or curved. . . . . 2. C. salicina
  bb. Ascospores straight. . . . . 3. C. myinda

1. Cryptodiaporthe aculeans (Schw.) Wehm. Figs. 189-192

On surface as pustules containing dense fascicles of elongate-cylindric ostioles erumpent through a disc which may be obliterated by the ostioles; perithecia spherical, 260-480 x 250-400 microns, with long slender necks; thickly clustered; no blackened zones in substratum; asci clavate, 47-65 x 5-8 microns; ascospores biseriate, long fusoid-ellipsoid, two-celled, hyaline, constricted at septum, 12-18 x 2.5-3 microns, usually with short hyaline appendage at each end.

In culture Wehmeyer (1933) noted two types of conidial stages produced; sporodochia with slender black stalks, 1.2 mm in length with a spherical capitate shiny black mass of conidia; conidia ovoid to fusoid, one-celled, dilute black, 6-9 x 2.5-3 microns; pycnidia in which are produced conidia ovoid to oblong-fusoid, one-celled, hyaline to light yellow, 5-8 x 2-3 microns.

On Rhus glabra L., Decorah, July, 1884, E.W. Holway (as Valsa aculeata Sz.); Woodman Hollow, June 27, 1958, L.H. Tiffany

On Rhus typhina L., McGregor, May 31, 1958, L.H. Tiffany

2. Cryptodiaporthe salicina (Curr.) Wehm. Figs. 197-200

On surface as numerous minute, conic pustules consisting of one or more ostioles surrounded by closely adherent and pustulate periderm; ostioles usually separately erumpent, often in clusters of 2-8, sometimes fused to form a small disc; no blackened zones within substratum; perithecia scattered singly or in small loose groups within bark cortex, spherical or somewhat flattened, 300-480 x 280-400 microns, walls membranous; asci clavate, 45-70 x 9-15 microns; ascospores biseriate, ellipsoid to inequilateral, blunt on ends, two-celled, but often tardily or faintly septate, hyaline, not constricted at septum, 15-28 x 3.5-7.5 microns.

In culture Wehmeyer (1933) obtained pycnidia containing conidia which were long cylindric to fusoid, straight to somewhat curved, 15-22 x 1.8-2.5 microns, sometimes becoming septate with age.

On Populus tremuloides Michx., Lake Okoboji, Aug. 6, 1956, L.H. Tiffany

On Salix alba L., Ames, April 9, 1958, L.H. Tiffany
On Salix babylonica L., Ames, June 4, 1958, L.H. Tiffany
On Salix sp., Ames, May 21, 1956, L.H. Tiffany; Lake Okoboji, July 21, 1956, L.H. Tiffany

American forms of C. salicina show a definite tendency to have spores which are longer, more strongly constricted at septum and more acute on the ends. Wehmeyer (1933) also suggests that the forms on Populus usually show spores with a greater range in size than do those on Salix and also a greater tendency for the perithecia to be clustered and the ostioles to be erumpent in small fascicles. These observations do not apply to all of our specimens.

Recently Butin (1957) has separated Cryptodiaporthe populea (Sacc.) Butin from C. salicina (Curr.) Wehm. Our material on both Populus and Salix corresponds to the latter species. Dothichiza populea Sacc. and Br. which Butin associated with Cryptodiaporthe populea has been reported from Iowa (Gilman and Archer, 1949) but no ascus stage has been found.

3. Cryptodiaporthe myinda (Cke. and Ell.) Wehm. Figs. 271-274

Numerous pustulate ruptures of the periderm through which short-cylindric ostioles are more or less erumpent either singly or in loose clusters; pustules often confluent longitudinally, forming long linear breaks in the periderm; perithecia spherical or somewhat flattened, 300-500 x 300-400 microns, loosely clustered or scattered in the unaltered bark cortex; surface of bark blackened where ostioles break through periderm, no dark zones in bark or wood; asci clavate, 50-65 x 13-15 microns; spores biseriate, broad fusoid-ellipsoid, two-celled, hyaline, constricted at septum, two-guttulate, 13-17 x 5-7 microns.

On Acer saccharum Marsh., Fort Defiance State Park, Aug. 7, 1958, L.H. Tiffany

10. Phragmodiaporthe Wehm.

Perithecia clustered, valsoid, entostromatic areas more or less definitely outlined by a blackened dorsal zone which is continuous between the pustules; asci clavate, 8-(4-?) spores; ascospores fusoid-ellipsoid to elongate, four- to many-celled, hyaline, or brown.

Conidia, where known, elongate-fusoid, one- to four-celled, hyaline, borne in clustered or labyrinthiform, enclosed locules within an entostroma.

A single species treated.

1. Phragmodiaporthe caryae (Peck) Wehm. Figs. 201-204

On surface as conic pustulate swellings, 0.5-2 mm in diameter with central cluster of stout-papillate ostioles or, where periderm is torn away, appearing as circular blackened discs; bark surface about ostioles blackened, blackening dips sharply into bark as lateral zone which may continue along wood surface; perithecia spheric to irregular from crowding, 300-600 x 200-500 microns, clustered, bounded by marginal zone; asci clavate, 85-150 x 19-21 microns; ascospores biseriate, long-fusoid, straight or slightly curved, hyaline, four-celled, slightly or distinctly constricted at septa, 27-44 x 5-7.5 microns.

Associated conidial stage has labyrinthiform to confluent irregular pycnidial cavities formed in entostromatic area of upper bark; conidia elongate, fusoid-clavate to oblong-cylindric, straight or somewhat curved, one-celled, or often two- to four-celled, hyaline, 35-43 x 3.5-4.5 microns.

On Carya ovata (Mill.) K. Koch, Ames, April 14, 1933, G.E. Alstatt; Ames, Sept. 30, 1955, L.H. Tiffany; Ames, May 5, 1957, L.H. Tiffany; Ames, Nov. 23, 1957, L.H. Tiffany; Boone County, May 11, 1958, L.H. Tiffany; Ledges State Park, June 12, 1955, L.H. Tiffany; Milford Woods, July 23, 1958, L.H. Tiffany

On Carya sp., Decorah, March, 1884 (Valsa apatela Ell. and Holw.); Wehmeyer (1941a)

Numerous collections of only the conidial stage have also been made.

11. Prosthecium Fres. emend. Wehm.

Pustulate erumpent ectostromatic disc well developed or practically absent. Very little entostromatic development. Asci stout-clavate, eight-spored. Ascospores usually rather large, cylindric-ellipsoid, bi- to triseriate in the ascus, many-celled, hyaline or brown, with cap-like or elongate to curved appendages at each end. Paraphyses broad, bandlike, evanescent.

Conidia many-celled, hyaline or brown, borne in open or, usually, in enclosed cavities within the ectostroma.

Key to the Species of the Genus PROSTHECIUM

a. Ascospores hyaline. . . . . 1. P. ulmi
aa. Ascospores brown
b. Appendages short and stout. . . . . 2. P. hapalocystis
bb. Appendages elongate. . . . . 3. P. corticale

1. Prosthecium ulmi Wehm. Figs. 205-207

On surface as small conical pustules 1.0-1.5 mm in diameter beneath the periderm, center of disc raised by compact cluster of black papilliform ostioles; perithecia small, 6-10 in pustule, 300-350 microns in diameter, entirely submerged in bark and not penetrating to wood; asci clavate to obovate, 75-150 x 25-60 microns, obscurely paraphysate; ascospores inordinate, slightly curved and subinequilateral, four-celled, 25-46 x 18-22 microns, hyaline at first, on maturity the center cells become olive-brown with lighter end cells, each with a cylindrical hyaline, straight or curved appendage, 12-20 x 5-6 microns.

Melanconium-like conidial stage obtained in culture by Wehmeyer (1941b); alpha conidia cylindric-ellipsoid to clavate, four-celled, hyaline, straight or slightly curved, 37-58 x 12.5-16 microns, exuded in pink to orange spore mass; beta conidia ellipsoid-oblong to fusoid, often tapered, one-celled, hyaline, 7-10.5 x 1-2 microns, produced in cavity within or on the surface of flatter, more effuse ectostromata, exuded in transparent yellow droplets.

On Ulmus americana L., Fort Defiance State Park, July 30, 1958, L.H. Tiffany

On Ulmus fulva Michx., Atlantic, Sept., 1933, Butler, No. 318; Wehmeyer (1941b)

This species was earlier reported as Pseudovalsa ulmi Wehm. by Gilman and McNew (1940).

2. Prosthecium hapalocystis (B. and Br.) Petr. Figs. 208-210

On surface as small flattened circular pustules 0.5-1 mm in diameter, scarcely visible minute central disc composed of few barely erumpent ostioles; more or less of an effuse hyaline entostromatic development about the perithecia, more strongly developed above but obliterated by development of the convergent ostioles; perithecia flattened, 250-500 x 200-250 microns, circinately arranged in upper bark; paraphyses broad, bandlike, soon evanescent; asci broad-clavate, 70-90 x 33-40 microns; ascospores irregularly triseriate, oblong-cylindric, ends broad and blunt, walls thick and gelatinous, constricted at septa on the inner margin of wall, straight or curved, three-celled, brown, 30-44 x 13-22 microns, with broad blunt cylindric straight or curved hyaline appendage at each end.

On Platanus occidentalis (Tourn.) L., Ames, March 22, 1957 L.H. Tiffany; Ames, Nov. 24, 1957, L.H. Tiffany; Ames, April 7, 1958, L.H. Tiffany

3. Prosthecium corticale (Schw.) Wehm. Fig. 211

On surface as numerous small flat circular pustules with minute central yellowish to dark-colored disc, 100-250 microns in diameter; periderm ruptured when young, in age surface of bark blackened by discharged spores; perithecia 250-500 microns, circinately arranged in surface bark just beneath periderm, adherent to periderm when that tissue removed; ostioles convergent and fused into disc which ruptures periderm; asci evanescent, broad-clavate, 85-110 x 40 microns; ascospores biseriate to triseriate, ellipsoid-fusoid, straight or curved, brown, mostly four-celled, occasionally two- or three-celled, somewhat constricted at septa, end cells of four-celled spores smaller, 46-70 x 13-20 microns, long tapered, straight to curved hyaline appendage at each end.

On Platanus occidentalis (Tourn.) L., Ames, Aug. 1954, L.H. Tiffany

Spores of P. corticale are usually four-celled, while those of P. hapalocystis are three-celled with a thick gelatinous wall and shorter blunter appendages.

12. Pseudovalsa Ces. and DeNot. emend. Wehm.

Ectostromata strongly developed, dark colored. Entostroma within the area of bark tissue, at first delimited by a blackened marginal zone. Ascospores more than two-celled, and two or three seriate within broad clavate asci. The conidial stage is referable to the form genus Coryneum.

Key to the Species of the Genus PSEUDOVALSA

a. Ascospores large and narrow, 45-75 x 5.5-11 microns. 1. P. longipes
aa. Ascospores smaller and broad
b. Entostromata black-brown, ascospores 35-56 x 13-18 microns. . . . . 2. P. umbonata
bb. Entostromata gray-black, ascospores 30-45 x 10-15 microns. . . . . 3. P. lanciformis

1. Pseudovalsa longipes (Tul.) Sacc. Figs. 44-47

On surface as small conical pustules, periderm irregularly ruptured, blackened disc with small papillate ostioles exposed but scarcely erumpent; perithecia 300-500 microns in diameter, clustered in isolate entostromatic area with rich proliferation of black-brown fungus tissue often outlined by blackened ventral zone; asci clavate, 100-180 x 17-25 microns; ascospores biseriate to irregularly triseriate, variable, elongate, cylindric-ellipsoid, one-celled and hyaline at first becoming four-celled and more curved to finally five- to eight- to ten-celled, brown, often constricted at septa, 35-80 x 6-11 microns.

Coryneum conidial stage obtained in culture by Wehmeyer (1941b); conidia curved, three- to eight-septate, dark brown, 47-104 x 10-14 microns; also beta conidia filiform, hamate, hyaline, one-celled, 8-15 x 1.0-1.5 microns. Same conidial stage referred by Tulasne to Coryneum kunzei.

On Quercus alba L., Holst State Forest, July 6, 1958, L.H. Tiffany
On Quercus imbricaria Michx., Albia, Oct. 1933, Burnett
On Quercus rubra L., Ledges State Park, Sept., 1933, Lee
On Quercus sp., Decorah, June 3, 1883, E.W. Holway
(Melanconis sigmoidea C. and E.)

The ascospores apparently mature slowly. Spores which were hyaline, five- to six-celled, with quite pointed end cells were common in our collections.

2. Pseudovalsa umbonata (Tul.) Sacc. Figs. 48-51

On surface as pustulate ruptures of periderm with central angular black carbonaceous disc 1-1.5 mm in diameter; ostioles merely punctate openings in roughened disc surface, entostroma truncate-conic, dark brown to black, finally crumbling away; perithecia 300-500 microns in diameter, 3 to 8 grouped in definitely outlined entostromatic area; ventral zone sharp; paraphyses broad, bandlike, soon evanescent; asci clavate, 150-160 x 28 microns; ascospores biseriate, oblong-ellipsoid, four- to six-celled, brown, 35-56 x 13-18 microns, not constricted, septa thin, each cell with a large guttula, end cells smaller, hyaline caplike portion of wall at ends.

Coryneum conidial stage; stromata blackish-cinereous; conidia formed on surface or in shallow cavities; conidia broadly ovate, four- to six-celled, dark brown, 40-50 x 19-22 microns; also short allantoid hyaline spermatia present.

On Quercus alba L., Amana, Oct. 5, 1957, L.H. Tiffany;
Fort Defiance State Park, Aug. 20, 1958, Judith Helin

3. Pseudovalsa lanciformis (Fr.) Ces. and DeNot. Figs. 251-254

On younger and smaller twigs as small circular to elliptic rather strongly pustulate black discs 0.5-1.5 mm in diameter, with closely adherent collar of periderm, on older stems as laterally elongate, fusoid to elliptic blackened discs, 1-5 x 0.5-1 mm, barely pustulate-erumpent through the thickened periderm; perithecia 400-600 microns in diameter, clustered within strongly developed gray-brown entostroma; entostroma often rather sharply outlined by ventral zone of darker color; ostioles circular, barely erumpent, often scarcely visible; asci clavate, with slightly thickened apical wall, 100-170 x 18-31 microns; ascospores biseriate, cylindric-ellipsoid, brown, six-celled, with faint cross walls and no constrictions, end cells somewhat smaller and end walls colorless, appearing as slightly differentiated caps, each cell with a rounded or angular guttula.

Coryneum conidial stage with slender filiform spermatia 9.5-13 microns in length, faintly curved and borne on erumpent stromata; conidia ovate-ellipsoid, four- to six-celled. Wehmeyer (1941b) produced similar Coryneum conidial stromata in culture which produced only conidia which were fusoid ellipsoid to clavate, tapering toward base, four- to six-celled, brown, and 31-45 x 13-15 microns.

On Prunus virginiana L., Ames, July 16, 1958, L.H. Tiffany

Host Index

Betula alleghaniensis
    Diaporthe beckhausii

Betula nigra
    Diaporthe beckhausii

Betula sp.
    Cryptospora betulae
    Melanconis decorahensis
    Melanconis stilbostoma

Caragena sp.
    Diaporthe medusaea

Carpinus caroliniana
    Diaporthe bakeri

Carpinus sp.
    Melanconis chrysostroma
        var. ellisii

Carya ovata
    Phragmodiaporthe caryae

Carya sp.
    Phragmodiaporthe caryae

Castanea dentata
    Melanconis modonia

Celtis occidentalis
    Diaporthe decedens

Chenopodium sp.
    Diaporthe arctii

Cladrastis lutea
    Diaporthe oncostoma

Cornus paniculata
    Diaporthe eres

Cornus sp.
    Apioporthe corni
    Diaporthe eres
    Melanconis corni

Corylus americana
    Apioporthe anomala
    Cryptospora ferruginea
    Diaporthe decedens
    Melanconis flavovirens

Corylus sp.
    Apioporthe anomala
    Cryptospora corylina
    Diaporthe decedens

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    Diaporthe eres

Erigeron sp.
    Diaporthe arctii

Fraxinus pennsylvanica
        var. lanceolata
    Diaporthe beckhausii
    Diaporthe eres
    Diaporthe medusaea

Glycine max
    Diaporthe phaseolorum
        var. batatatis
    Diaporthe phaseolorum
        var. sojae

Juglans cinerea
    Diaporthe spiculosa
    Melanconis juglandis

Juglans nigra
    Diaporthe spiculosa
    Melanconis juglandis

Juglans regia (?)
    Diaporthe medusaea

Juglans sp.
    Diaporthe spiculosa

Lonicera tatarica
    Diaporthe eres

Ostrya virginiana
    Melanconis chrysostroma
        var. ellisii
    Melanconis ostryae

Parthenocissus sp.
    Cryptosporella viticola

Phalaris arundinacea
    Diaporthe phaseolorum

Physocarpus opulifolius
    Diaporthe eres

Platanus occidentalis
    Prosthecium corticale
    Prosthecium hapalocystis

Populus tremuloides
    Cryptodiaporthe salicina
    Melanconis occulta

Populus sp.
    Melanconis apocrypta

Prunus americana
    Diaporthe prunicola

Prunus hortulana
    Diaporthe padi
    Diaporthe prunicola

Prunus serotina
    Diaporthe pruni

Prunus virginiana
    Diaporthe padi
    Diaporthe pruni
    Pseudovalsa lanciformis

Prunus sp.
    Diaporthe eres
    Diaporthe prunicola

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    Diaporthe eres

Quercus alba
    Apioporthe macrospora
    Diaporthe leiphaemia
        var. raveneliana
    Diaporthe spiculosa
    Diaporthe taleola
    Pseudovalsa longipes
    Pseudovalsa umbonata

Quercus imbricaria
    Pseudovalsa longipes

Quercus macrocarpa
    Diaporthe taleola

Quercus rubra
    Pseudovalsa longipes

Quercus sp.
    Cryptospora albofusca
    Diaporthe eres
    Diaporthe leiphaemia
        var. raveneliana
    Diaporthe taleola
    Pseudovalsa longipes

Rhamnus cathartica
    Diaporthe eres

Rhus glabra
    Cryptodiaporthe aculeans

Rhus typhina
    Cryptodiaporthe aculeans

Rhus sp.
    Diaporthe peckii

Ribes missouriense
    Diaporthe strumella

Ribes sp.
    Diaporthe strumella

Robinia pseudoacacia
    Diaporthe oncostoma

Salix alba
    Cryptodiaporthe salicina

Salix babylonica
    Cryptodiaporthe salicina

Salix nigra
    Diaporthe tessella

Salix sp.
    Cryptodiaporthe salicina
    Diaporthe tessella

Sambucus canadensis
    Diaporthe megalospora
    Diaporthe sociabilis
        var. sambuci

Scirpus sp.
Phomatospora bostryosphaerioides

Smilax sp.
Melanconis smilacis

Tilia americana
Cryptospora tiliae
Diaporthe eres
Diaporthe spiculosa
Diaporthe tiliacea
Melanconis juglandis var. tiliae

Ulmus americana
Apioporthe apiospora
Diaporthe eres
Diaporthe medusaea
Diaporthe otthii
Diaporthe padi
Melanconis acrocystis
Melanconis sudans
Prosthecium ulmi

Ulmus fulva
Prosthecium ulmi

Ulmus sp.
Apioporthe apiospora
Diaporthe eres
Diaporthe medusaea

Viburnum lentago
Cryptosporella lentaginis
Diaporthe viburni

Viburnum sp.
Diaporthe viburni

Vitis labrusca
Diaporthe medusaea

Vitis sp.
Cryptosporella viticola

Xanthoxylum americanum
Diaporthe medusaea

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PLATE 1

Figs. 1-5. Cryptospora corylina
1. Perithecia. 2. Habit. 3. Perithecium. 4. Ascus. 5. Ascospores.

Figs. 6-8. Cryptospora femoralis
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Figs. 13-15. Cryptospora tiliae
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19. Habit. 20. Pycnidium. 21. Conidia. 22. Ascus (after Shear, 1911).

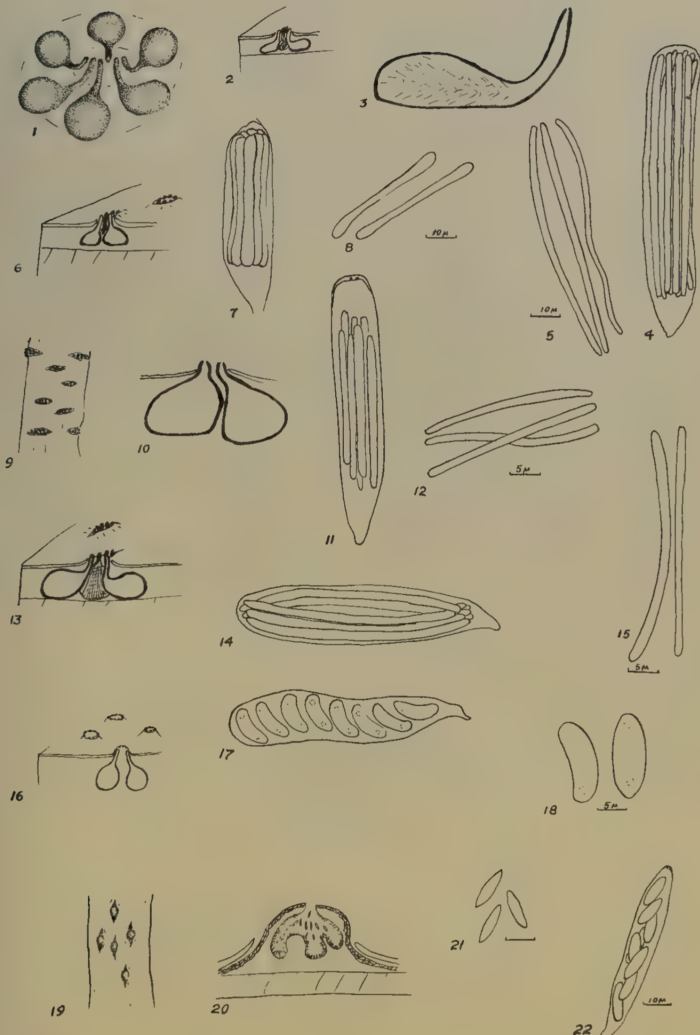
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PLATE 2

Figs. 23-26. Diaporthopsis angelicae
23. Habit. 24. Perithecium. 25. Ascus. 26. Ascospores

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27. Habit. 28. Perithecium. 29. Ascus. 30. Ascospores (1-celled).
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Figs. 40-43. Apioporthe apiospora
40. Habit. 41. Perithecia. 42. Ascus. 43. Ascospores.

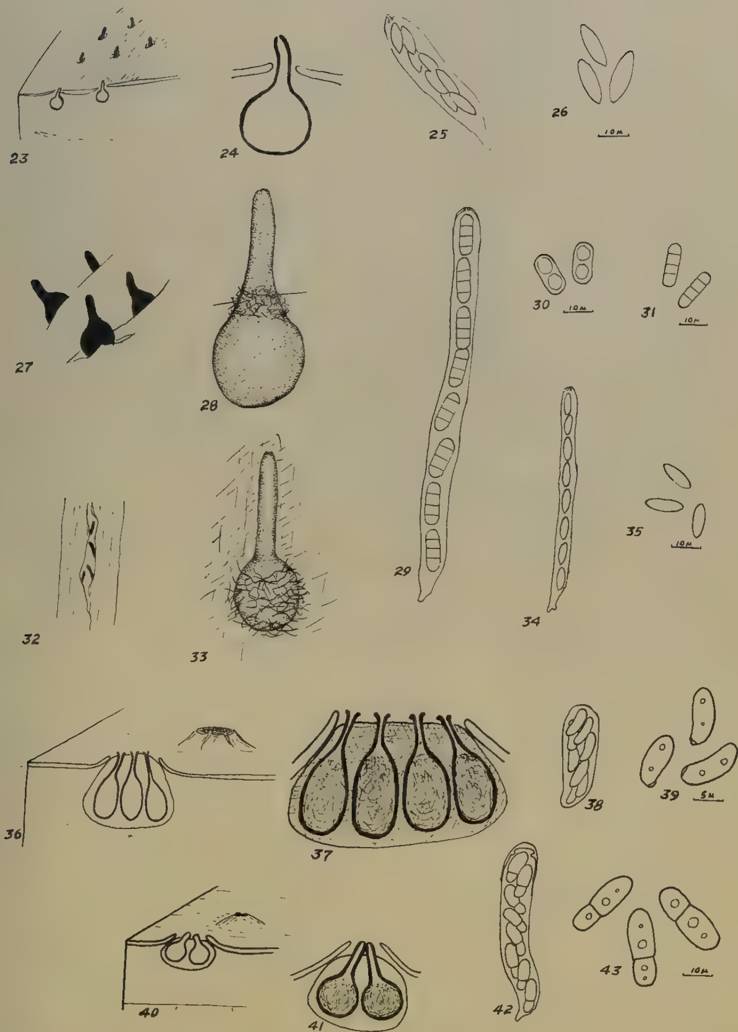
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PLATE 3

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56. Habit. 57. Ascus. 58. Ascospores.

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59. Perithecia. 60. Ascus. 61. Ascospores.

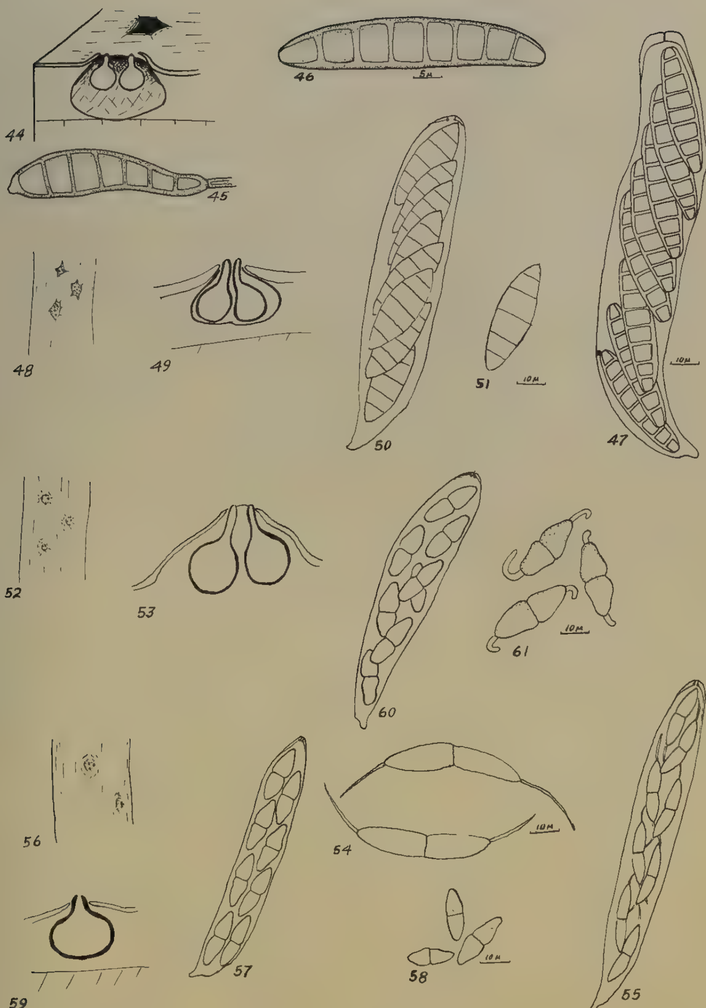
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PLATE 4

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62. Habit. 63. Perithecia. 64. Ascus. 65. Ascospores.

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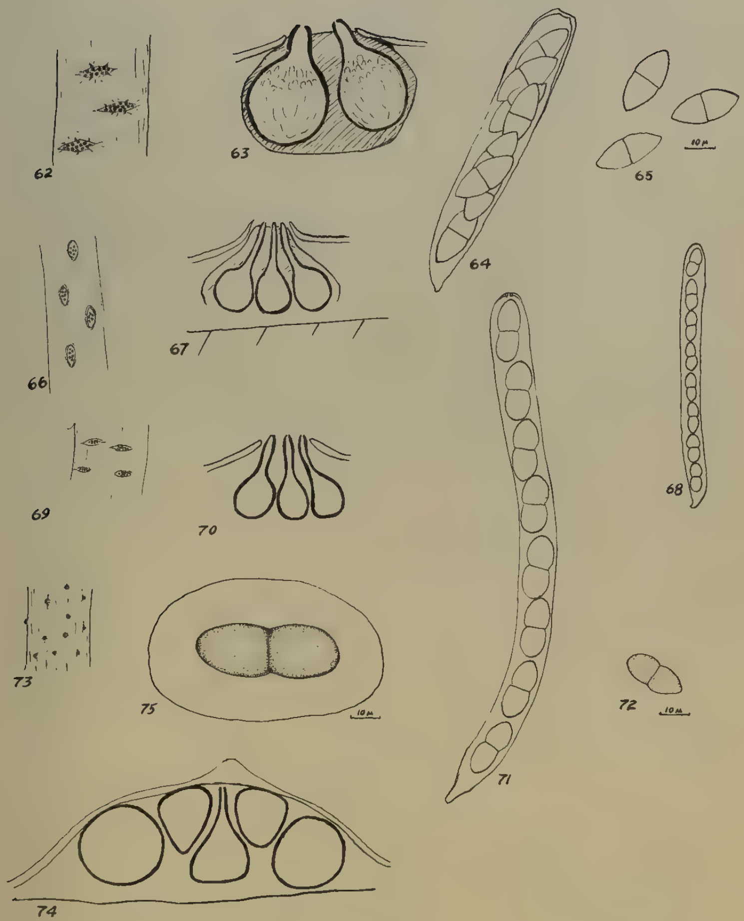
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PLATE 5

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92. Ascus. 93. Ascospores.

Figs. 94-96. Melanconis juglandis
94. Perithecia. 95. Ascus. 96. Ascospores.


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PLATE 6

Figs. 97-100. Diaporthe phaseolorum var. sojae
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109. Habit. 110. Perithecia. 111. Ascus. 112. Ascospores.

Figs. 113-116. Diaporthe eres
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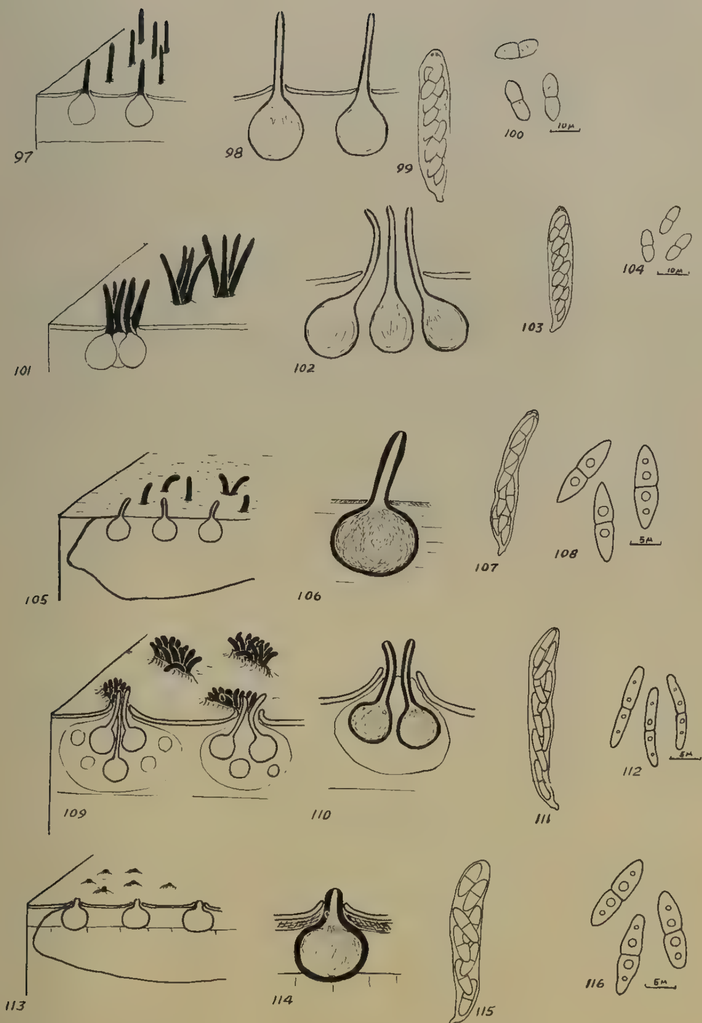
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PLATE 7

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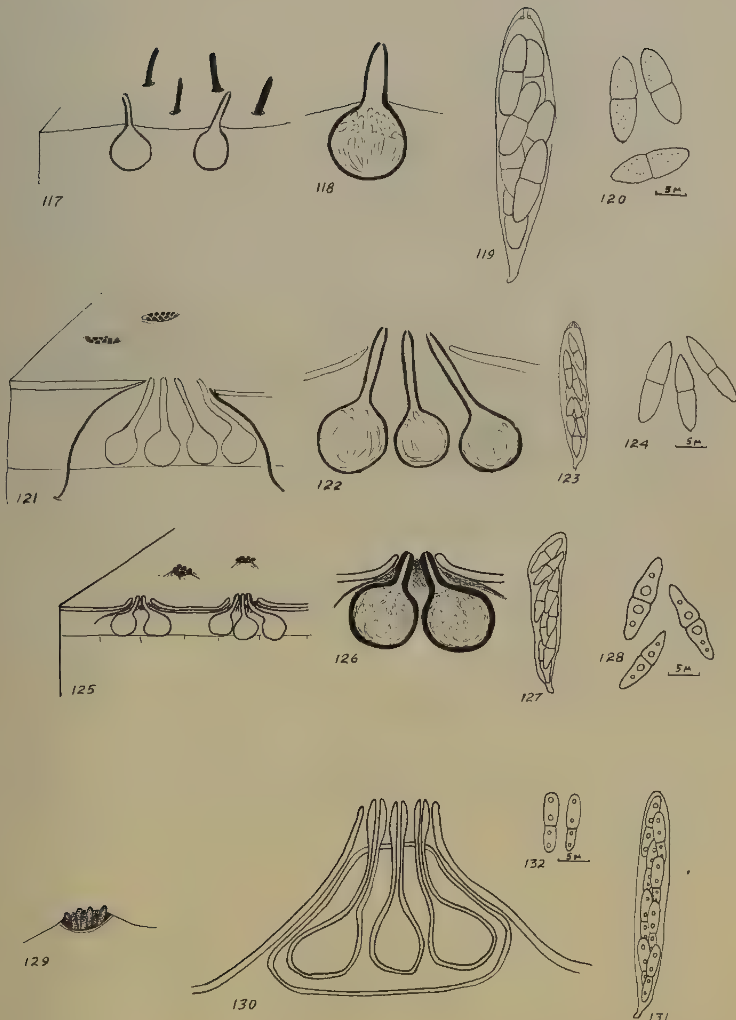
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PLATE 8

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153. Beta conidia. 154. Ascospores.

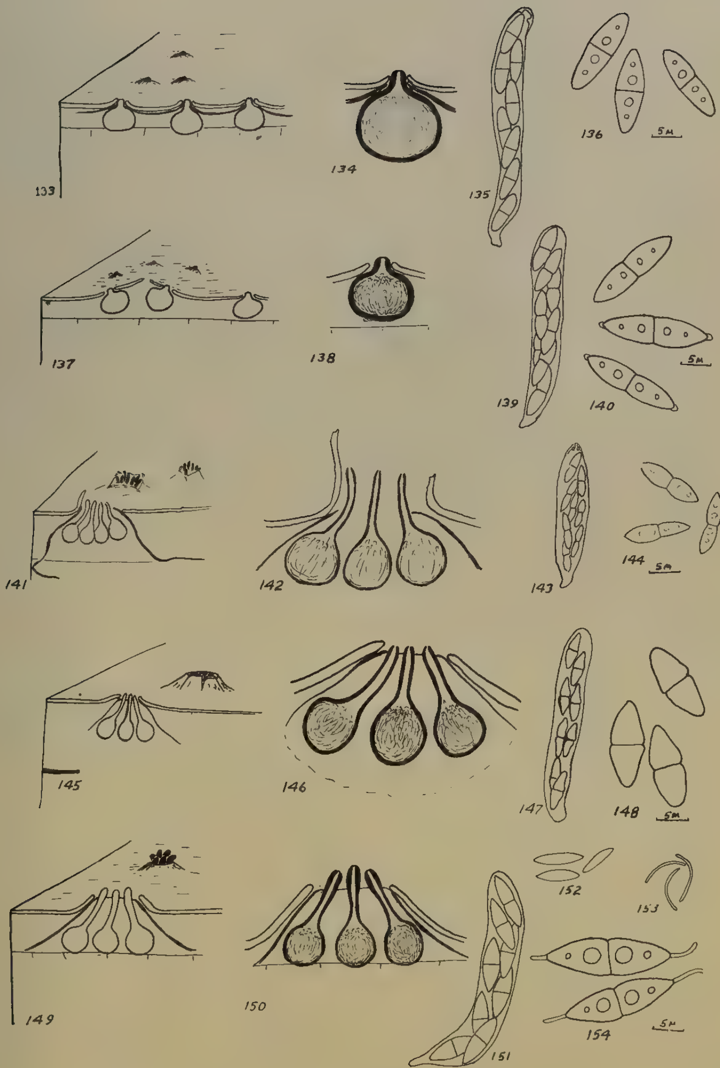
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PLATE 9

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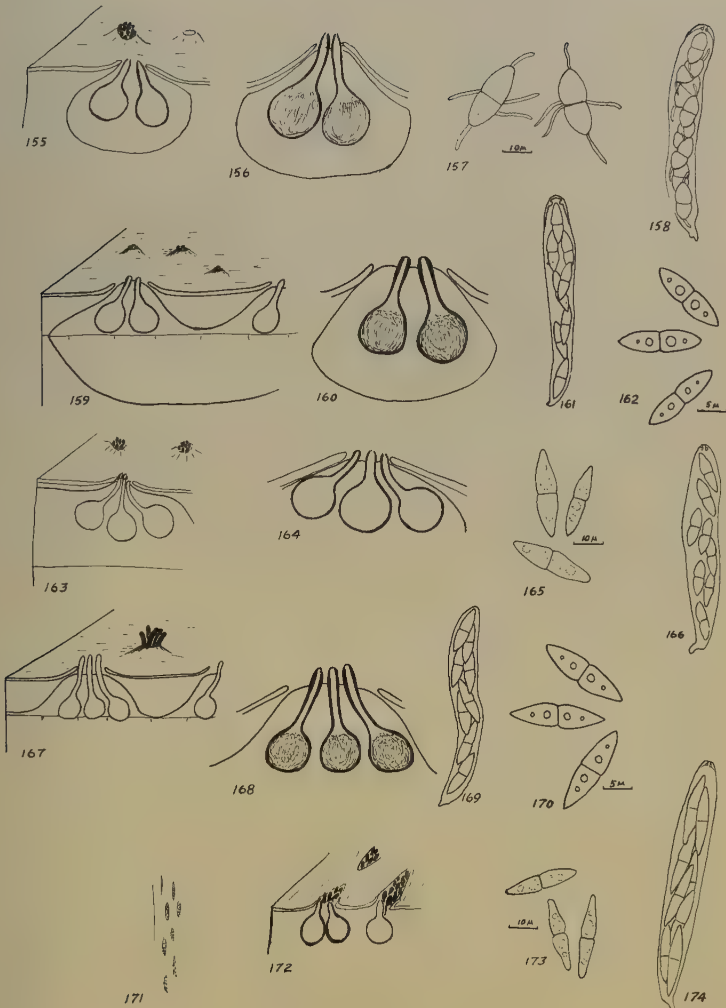
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PLATE 10

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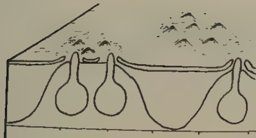

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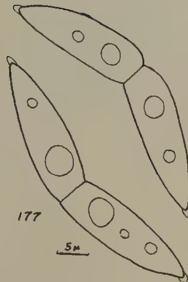

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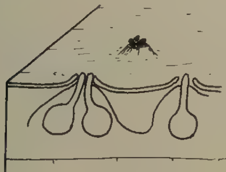

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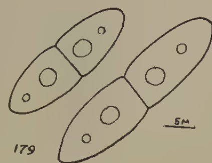

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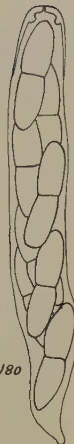

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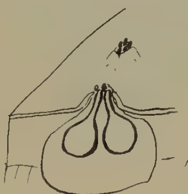

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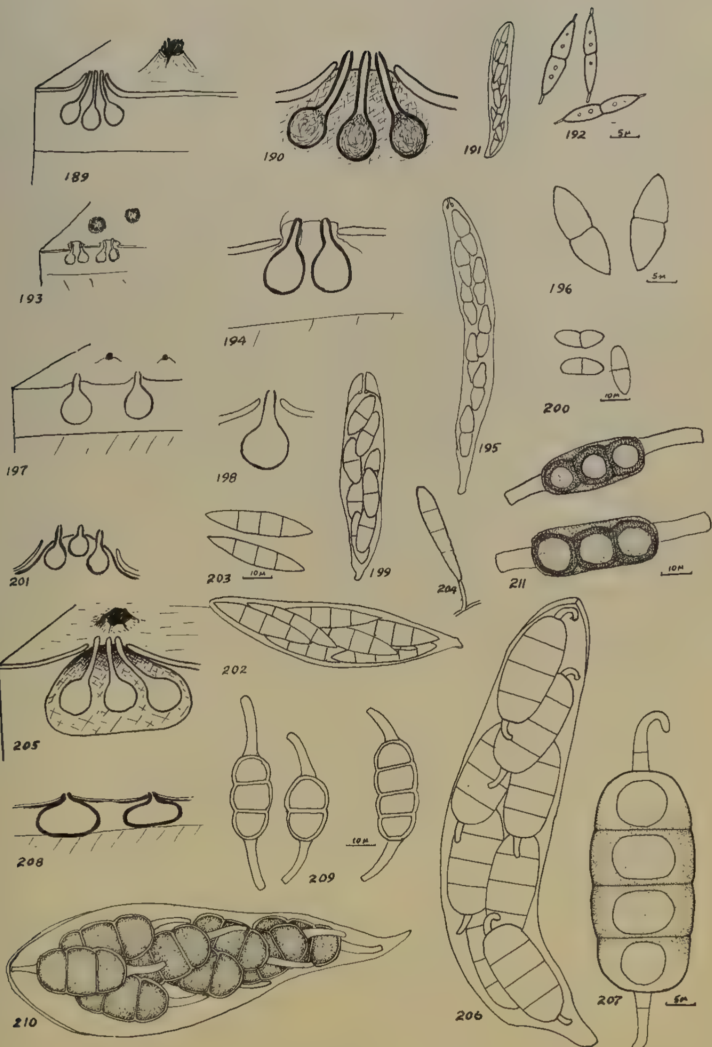
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212. Habit. 213. Perithecia. 214. Ascus. 215. Ascospores.

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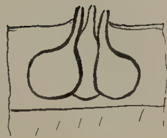
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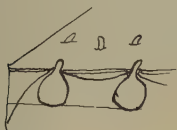
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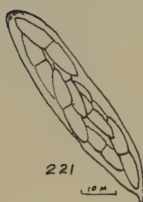

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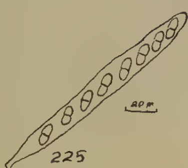

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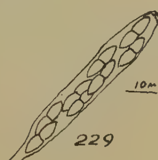

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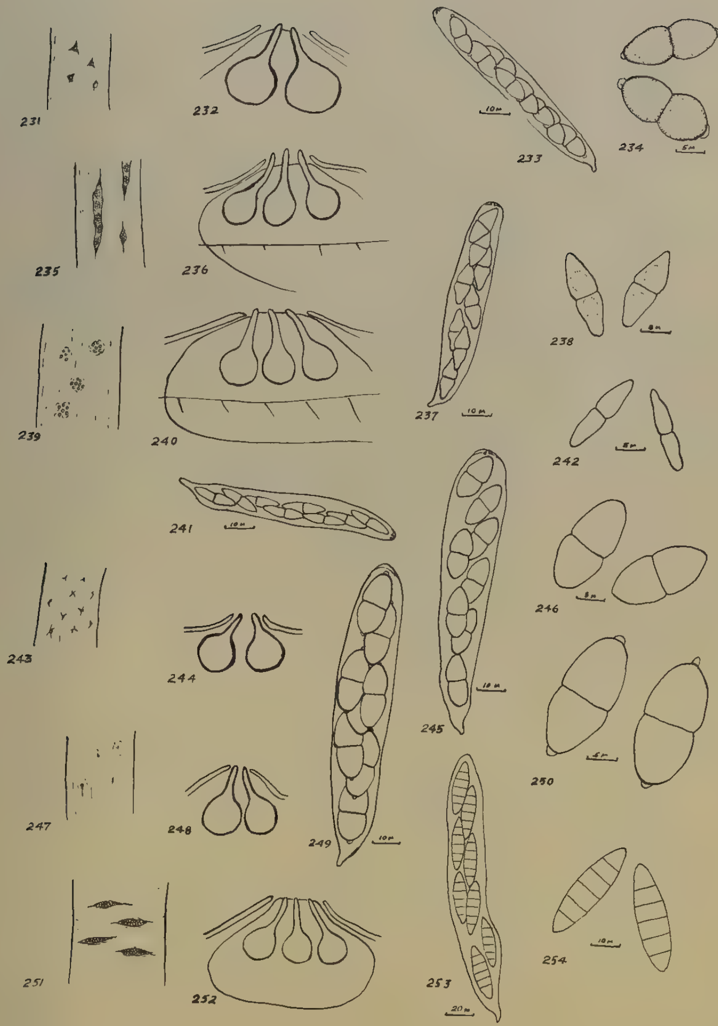
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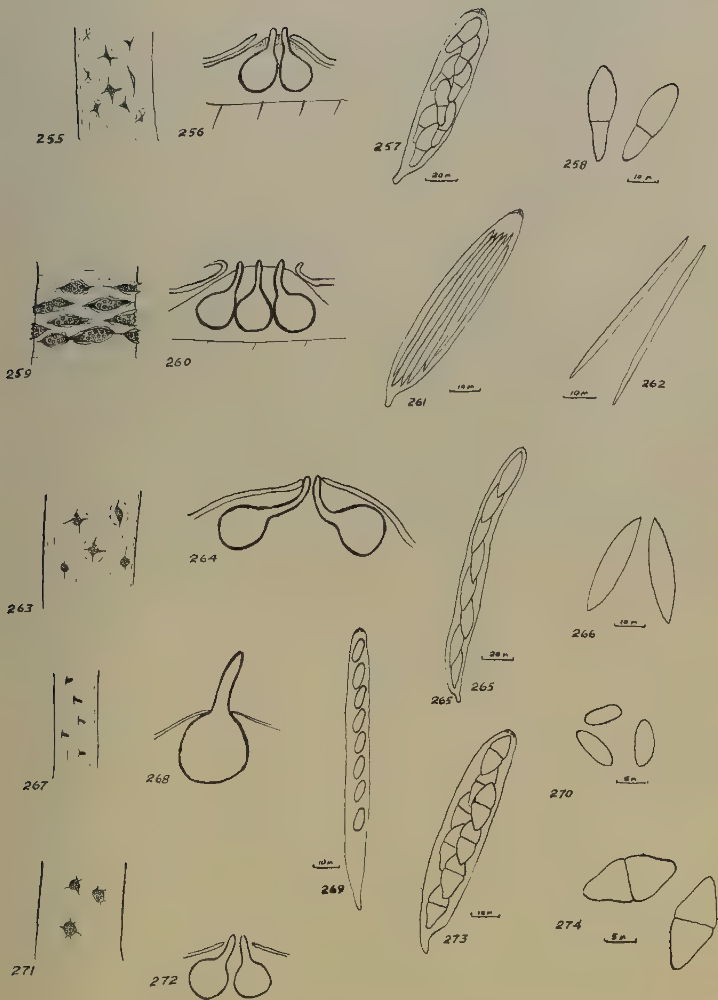
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PRODUCTION OF BOTTOM FAUNA IN THE PROVO RIVER, UTAH[1]

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INTRODUCTION

Life is precarious in mountain streams and a fine degree of fitness is necessary for those plants and animals found there. Constantly changing from day to day, from month to month, and from season to season, stream conditions offer a highly unstable and complicated environment. Man has further accentuated the instability of this environment by his various activities. The seasons often bring sudden changes in volume and velocity of water that wipe out whole aquatic populations in a short time. The specialized conditions restrict the number of animal and plant species very markedly. Indeed, in our best trout waters—clear, cold, mountain streams, the larger aquatic plants, upon which many aquatic invertebrates depend for their livelihood, are practically eliminated by the current. The biota is further limited to species that are either strong swimmers or have special structural adaptations for clinging.

A striking feature of mountain streams is the rapid and abrupt change of habitats. In a small area may be found all the extremes from a vertical to a horizontal flow of water; from shallow, placid stretches to deep, stone-lined pools. Animals and plants assemble in small communities or biotic islands, often separated by barren areas kept uninhabited by the severity of the current.

The Provo River is a typical mountain stream of the Intermountain region and is well known for its trout fishing. Field investigations on this river were initiated by the writer on September 15, 1946 and continued to May 28, 1949.

The general purpose of this study was to obtain a quantitative and qualitative measure of the stream bottom invertebrates of the river as potential sources of food for the trout populations present. Another objective was to learn something about the life histories, habits, and adaptations of the invertebrate inhabitatns of the stream. Various physical, chemical, and biotic factors which influence the productivity of a river were measured. Finally, since very few investigations have

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[1] Part of a thesis filed in partial fulfillment of the requirements for the degree of Ph.D. at Iowa State College. This project was sponsored by the Research Committee of the University of Utah and the Utah Fish and Game Department.

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dealt with conditions existing in streams during the winter season, special attention was given to the determination of the ecological changes occurring during that time of the year.

METHODS

In order to select sampling stations which would be representative of the different sections of the stream the author spent the first ten months of study conducting seasonal reconnaissance surveys of the entire river. Information concerning the geology and topography of the drainage basin was gathered. Collections of fauna and flora were made from as many different habitats as could be found. Changes in water level and course brought about by the multiple uses of the river for irrigation, power, and recreation were determined.

In making qualitative collections of the fauna in the stream a Needham hand screen sampler was found to be most useful. Hand picking of nymphs and larvae from rocks, debris, and vegetation was also used effectively on many occasions. Adults were picked from bridges, rocks, and buildings or by sweeping the vegetation. The specimens collected were preserved in vials of 80% ethyl alcohol and were later identified to genus or species.

Following this reconnaissance phase of the project, nine major stations were selected in typical average sections of the stream from the headwaters to its mouth. Chemical, physical, and biological data and samples were taken at each station on a weekly basis from June to September, 1947. Sampling and collecting of data were conducted on a monthly basis during the following twenty-one months of the study.

The direction of flow and the stream gradient were determined by use of a surveyor's transit and stadia rod. The approximate altitude at each station was obtained by reference to United States Geological Survey maps of the region. Average widths and depths were determined by actual measurements and by reference to stream bottom profiles and staff gauges which were constructed at the beginning of the project. Stream velocities were measured by timing the passage of floats over one hundred foot sections at each station. The volume of flow was determined by using the formula given by Embody (1927). The turbidity and color of the water were obtained by means of a United States Geological Survey turbidity rod and glass color disk outfit. Air and water temperatures were measured by standardized chemical thermometers.

All water samples for chemical analysis were taken below the stream's surface with a 1.5 liter Kemmerer water bottle. Hydrogen ion concentrations were determined colorimetrically with a La Motte set, using cresol red or bromthymol blue as indicators. The unmodified Winkler method as outlined in Standard Methods of Water Analysis (1947) was used for the determination of dissolved oxygen. Free carbon dioxide values were obtained by titrating 100 cc. samples of water with N/44 sodium hydroxide using phenolphthalein as an indicator. Phenolphthalein and methyl orange alkalinities were obtained by titrating a 100 cc. sample of water against N/50 sulfuric acid using phenolphthalein and methyl orange as indicators. The electrolytic content of the water was determined by means of a Leeds and Northrop Soil Meter connected to

a Heathkit Oscilloscope in such a way as to give equivalent electrical resistance in ohms at 60°F.

Quantitative bottom samples were taken by use of a Surber square foot sampler (Welch 1948, pp. 321-322). From one to three samples were taken at random from each station each inspection date. The organisms were transferred to gallon bottles, and taken to the laboratory where they were separated to orders, counted, and preserved in 80 per cent alcohol for future study. All specimens collected were later studied under the binocular microscope, counted, and identified to genus or species. Volumetric measurements of each genus or species were made using the liquid displacement method.

GENERAL DESCRIPTION OF THE PROVO RIVER

The Provo River arises in the high Uinta Mountain area of Northeastern Utah and runs approximately 72 miles in a southwestern direction to Utah Lake (Fig. 1). The Main Fork originates in several mountain lakes and receives several sizeable tributaries during the first fourteen miles of its course. In the upper section of the river there are several conspicuous falls and cascades and the gradient is over 400 feet during the first mile. Ten miles from its source it pauses in its downward rush and passes placidly through a series of beaver ponds, only to resume its turbulent course. At Stewart's Ranch the Main Fork is joined by the South Fork which makes a profound change in the chemical nature of the water. In its descent from its source to the junction with the South Fork, about 2800 feet in seventeen miles, the river passes through an area composed largely of relatively insoluble quartzite mixed with a small amount of shale. The South Fork originates in the Wolf Creek Pass area seven miles southeast of Stewart's Ranch and has a gradient of 2400 feet in that distance. Its drainage area consists largely of Mississippian limestone. The river formed by the junction of the two forks flows southwest approximately seventeen miles to Deer Creek Reservoir. In this section the river is less turbulent than above and has a drop of only 1600 feet (Fig. 2).

Deer Creek Reservoir is approximately eight miles long and the water undergoes many physical and chemical changes in passing through. After leaving the reservoir the river plunges through alternating series of placid pools, gentle rapids and turbulent twisting cascades in Provo Canyon. During its descent it receives waters from three streams, Deer Creek, Aspen Grove Creek, and Vivian Park Creek. These streams pass through geological formations consisting of about 80 per cent limestone, and the waters which they contribute are highly aklaline. The gradient of the river from the base of the reservoir to the foothills beyond the mouth of the canyon is about 600 feet for the fifteen mile section. As the river reaches Utah Valley its gradient gradually reduces and the rocky bed gives way to gravel and in some places sand. The water gets warmer and finally quite sluggish before entering Utah Lake.

The source of the water in the river is chiefly melting snow and springs. Ordinarily there is very heavy snowfall over the entire area and the river and its tributaries are decidedly torrential during the early thaws. The basic flow derived from springs is fairly constant from July

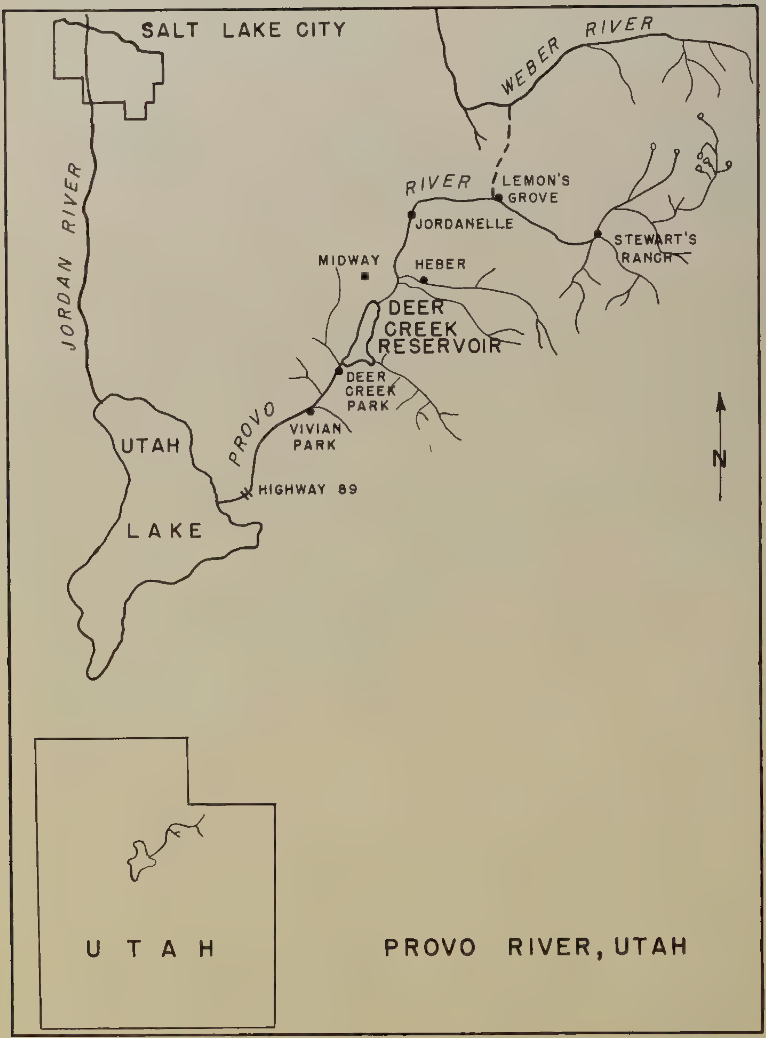

Fig. 1. Map of Provo River showing principal research areas, and an inset showing location of the River in Utah.

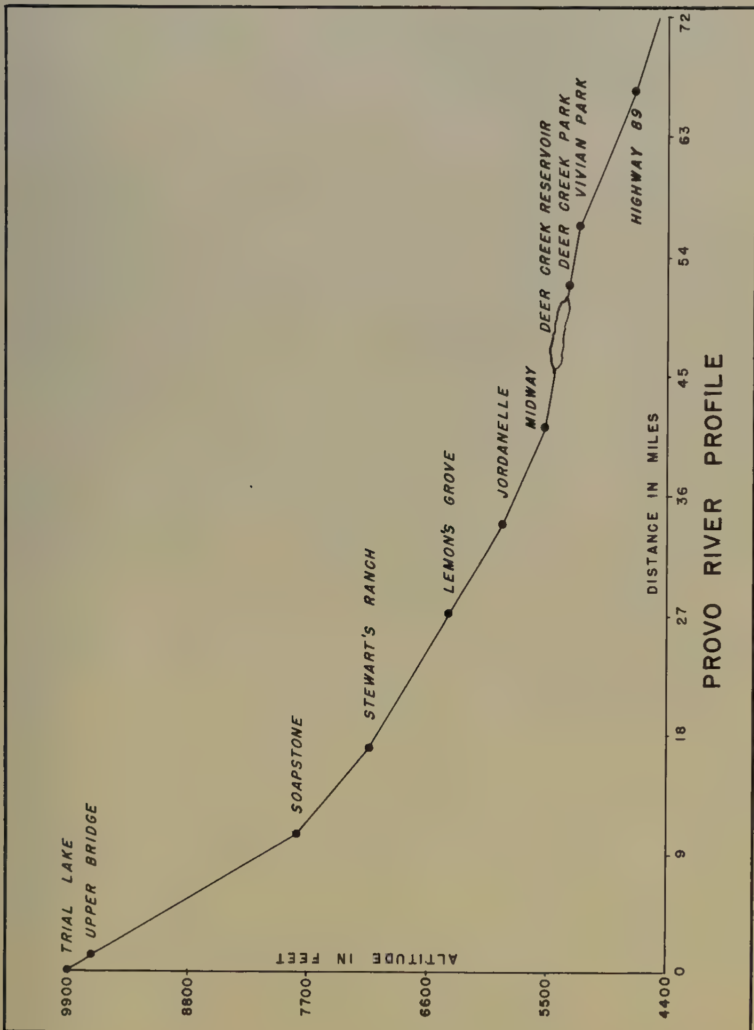

Fig. 2. Profile of the Provo River.

to April, but the melting snows of spring add to the basic flow and produce a marked rise of 40 to 150 per cent from April to June. In addition, sudden floods of irregular proportions may occur during the cloudburst period from July to mid-September.

Every step in the downward progress of the river from the Uinta Lakes to the basin bottom is accompanied by variations in temperature, light, soil, soluble minerals, topography, aspect, and many other factors of the environment. In its passage downward the river passes through several different ecological zones. In the Provo River drainage area, the higher mountains (8, 000 to 11, 000 feet) bear spruce-fir forests as a dominant type of vegetation, except where the higher mountain peaks project above timberline. The blue spruce, Picea pungens, and white fir, Abies concolor, are the dominant trees of the lower conifer belt, but from 9, 500 feet to timberline these trees are replaced by Engelmann spruce, Picea engelmanni, and alpine fir, Abies lasiocarpa. The Douglas fir, Pseudotsuga taxifolia, is common throughout the entire conifer belt. Interspersed with the spruce-fir stands in many places in the Uinta Mountains are thick, single-edged stands of lodgepole pine, Pinus contorta murrayana. Also common are aspen, Populus tremuloides, forests which occur between 7, 000 and 9, 500 feet elevation. Both of the latter types are unable to grow in the shade and are therefore only temporary forests, being replaced by spruce and fir in the ecological succession. These forests aid the storage of snow by holding and shading, thus delaying the melting period. They also furnish litter and humus for the soil, which increase the absorbing capacity of the soil and add to the underground water supply for springs.

The lower mountain slopes (6, 000 to 8, 000 feet) are robed with deciduous chaparral, chiefly oak brush, Quercus gambelii. This furnishes an excellent source of forage for livestock grazing in the summer. Overgrazing has resulted in depletion of the vegetative cover in some areas along the watershed, reducing the absorbing capacity of the soil, producing scouring floods, and adding silt to the stream.

The foothill slopes (5, 000 to 7, 000 feet) bear pigmy forests of juniper, Juniperus spp., and pinyon pines, Pinus spp., widely spaced with root systems several times as large as the crowns. The open, well-drained valleys (5, 000 to 6, 000 feet) along the course of the river are largely utilized for agriculture. Much grassland exists, supporting sizeable herds of dairy cattle. Fruit trees, wheat, and alfalfa are common, requiring diversion of some water from the river for irrigation purposes. Along the streamside at higher elevations are found numerous willows, Salix spp., and at lower elevations, birch, Betula fontinalis, alder, Alnus tenuifolia, cottonwood, Populus deltoides, maple, Acer glabrum, and willows, Salix spp. abound.

PHYSICAL AND CHEMICAL DATA

The various physical and chemical factors which affect the life in streams are all interrelated and changes in any one factor may influence all of the others. Most of the differences in these factors which occurred in the Provo River could be largely attributed to seasonal and altitudinal effects.

Table 1. Comparison of velocity and volume of flow at various stations in the Provo River, Utah, June 17, 1947, to May 30, 1949.

Stations	Elevation in feet	Velocity (ft./sec.) Min.	Velocity (ft./sec.) Max.	Volume (cu.ft./sec.) Min.	Volume (cu.ft./sec.) Max.
Upper Bridge	9,500	1.5	7.5	11.3	155.2
Soapstone Ranger Sta.	7,750	0.9	9.6	25.4	596.1
North Fork	7,500	1.1	9.2	6.7	435.5
Stewart's Main Fork	7,100	1.45	7.6	32.2	585.5
Stewart's South Fork	7,100	1.8	4.9	27.2	98.3
Lemon's Grove	6,400	2.2	7.2	84.8	499.4
Jordanelle	5,900	0.33	9.2	1.6	1,062.8
Heber-Power House	5,800	0.82	6.7	5.5	109.4
Heber-Midway	5,500	0.64	8.2	20.0	2,250.0
Deer Creek Park	5,300	2.05	5.9	233.2	858.4
Vivian Park	5,200	1.5	9.0	67.5	2,290.0
Deer Creek	5,300	3,4	5.7	17.2	60.9
Aspen Grove Creek	5,250	2.6	8.1	8.7	217.2
Vivian Park Creek	5,200	3.9	5.7	47.1	78.6
Highway 89	4,489	1.1	6.3	25.4	1,147.1

Most mountain streams are characterized by great variations in flow, and the Provo River is no exception (Table 1). During the present study the flow ranged from a trickle of only 1.6 cubic feet per second on September 3, 1947 at Jordanelle to a torrent of 2,290 cubic feet per second on May 22, 1948 at Vivian Park. In general, the stream was lowest during the fall and winter, and highest during the spring and early summer. Greatly decreased runoff in the hills and mountains correlated with low precipitation, and diversion of considerable water from the river for power and irrigation purposes resulted in very low flows during the late summer, fall, and winter.

Seasonal variations in velocity correlated closely with the changes in volume. The lowest velocity recorded was 0.33 feet per second also taken at Jordanelle on September 3, 1947 while the highest reading of 9.6 feet per second was taken at Soapstone Ranger Station on May 29, 1948. The reading at Vivian Park on May 22, 1948 was also high with 9 feet per second being recorded.

So few measurements of turbidity and color were made with the U. S. Geological Survey set that no accurate statistical correlation can be made between seasonal flows and turbidity and color. However, an obvious relationship usually existed between the height of the stream and the amount of silt in suspension. When the flow was high in the spring and early summer the presence of silt in the water could readily be detected with the eye. The highest turbidity recorded with the measuring rod was 90 ppm on May 5, 1949 at the Heber-Midway Station.

Table 2. Comparison of important physical characteristics at different stations in the Provo River, Utah, June 17, 1947, to May 30, 1949.

Stations	Average gradient per mile (in feet)	Average width (in feet)	Average depth (in feet)	Average annual water temp. (°F)
Upper Bridge	132	24	0.67	52
Soapstone Ranger Station	108	32	1.4	55
North Fork	132	27	0.58	52
Stewart's Main Fork	95	35	1.17	47
Stewart's South Fork	68	24	1.0	44
Lemon's Grove	77	42	1.25	51
Jordanelle	32	57	1.25	51.8
Heber-Power House	33	9	1.58	51.8
Heber-Midway	33	88	1.7	52.6
Deer Creek Park	61	88	2.38	49
Vivian Park	43	75	1.58	51.1
Deer Creek	100	10	0.62	47
Aspen Grove Creek	100	20	0.62	47
Vivian Park Creek	100	18	0.75	47
Highway 89	61	83	0.92	55.3

This contrasted with readings of less than 7 ppm taken during low flows at all of the stations. The maximum color recorded was 60 at Soapstone Ranger Station on May 28, 1949. Color readings of 40 to 50 were also made at all other stations during that month. The color, characteristic of all of the stations at other seasons, was less than 10 with zero readings being common.

One characteristic of a mountain stream is the lack of thermal and chemical stratification such as may be found in lakes, ponds, and sluggish rivers. The waters are constantly churning and mixing. The average annual stream temperature for the period 1947-1949 was 51.8°F (Table 2). The highest temperature recorded was 71°F on August 6, 1947 at the Heber-Midway Station. This was correlated with very low flow (less than 20 c.f.s.), low velocity (0.64 feet per second), and a comparatively high air temperature (78°F). The lowest water temperature found was 32.2°F, recorded several times from December to February at Stewart's Ranch in the Main Fork of the river. This low temperature was characteristic of winter conditions throughout the entire upper Main Fork.

Dissolved oxygen concentrations were quite similar in all sections of the river and never fell below 6.8 ppm. That low value was recorded on August 12, 1947 at Heber-Midway. There was a seasonal trend

in the oxygen concentrations but this was nothing more than could be anticipated on the basis of changes in water temperatures. In the series of 267 determinations made during the study, oxygen ranged from 73 to 140 per cent saturation, with 155 determinations being below and 112 readings above saturation.

The amount of free carbon dioxide in solution in the river at all times was low. As could be expected the constant agitation of the water prevented the retention in solution of more than a small amount. The highest reading of 8.0 ppm was made on the Main Fork of the river at Stewart's Ranch on February 26, 1949. This followed a period in which the stream was covered by a solid ice sheet for two months. The second highest reading of 5.5 ppm was made early in the morning on August 27, 1948 at Heber-Midway from a pool heavily populated with algae. For 127 of the 267 determinations no free carbon dioxide could be detected. Absence of free carbon dioxide was characteristic of all of the lower stations during most of the study period.

The waters of the Provo River were always alkaline except during the spring runoff, and then, only the upper 17 miles of the stream displayed a neutral or slightly acid reaction (Table 3). Data collected from the Upper Bridge, Soapstone Ranger Station, and North Fork from May to June in 1947, 1948, and 1949 showed pH determinations of 6.8 and 6.9. In the late summer, fall, and winter tha pH range at those stations was between 7.1 to 7.3. The river at the other stations above Deer Creek Reservoir was always on the alkaline side, with the water becoming decidedly more alkaline as the flow subsided from the high spring level. Below the reservoir this trend was not so evident due to the stabilizing influence of the lake conditions above. The higher alkalinity of the stream in the fall and winter was primarily due to increased dissolved mineral content resulting from decreased volume of flow, and to increased photosynthetic activity associated with the buildup of the algal population.

The amounts of bound carbon dioxide showed considerable variation from a low of 4.5 ppm on May 28, 1949, at the Upper Bridge to a high of 222 ppm at Heber-Midway on August, 1948. Much of this variation is attributable to differences in the nature of the surface soil and rocks over which the stream flows as it descends to the valley, but the age of the water is also an important factor to consider. According to Pennak (1943) there is a correlation between the bound carbon dioxide content and stream flow, the theory being that during rains or rapid melting of snows, the water passes over the surface of the ground in rivulets and consequently does not dissolve as much carbonate as it would if it moved more slowly. According to Shelford (1925) the salt content of a water is determined by its age, i.e., the time since it melted from snow or ice or fell as rain, and by the kind of materials over which it flows. Observations in the Provo River showed that there was a definite relationship between stream flow and bound carbon dioxide. The lowest methyl orange and phenolphthalein alkalinity readings at all stations were taken during periods of high flow in the spring, while the highest values were associated with low flow conditions in the late summer and fall. While geographical conditions and age of the water were the most important factors involved in producing the variations in alkalinity observed, other seasonal factors also played a part. When the early melting snows and rains

Table 3. Hydrogen ion concentration and carbonate alkalinity at various stations of the Provo River, Utah, June 17, 1947, to May 30, 1949.

Stations	pH Min.	pH Max.	Carbonate Alkalinity in ppm Min.	Carbonate Alkalinity in ppm Max.	Carbonate Alkalinity in ppm Mean
Upper Bridge	6.8	7.3	4.5	10.0	7.8
Soapstone Ranger Station	6.9	7.3	10.0	34.0	17.0
North Fork	6.9	7.4	8.0	16.0	12.6
Stewart's Main Fork	7.0	7.7	18.0	30.0	25.0
Stewart's South Fork	7.6	8.3	139.0	160.0	144.0
Lemon's Grove	7.5	8.5	42.0	140.0	97.0
Jordanelle	7.5	8.5	65.0	124.0	96.0
Heber-Power House	7.7	8.1	63.0	102.0	89.0
Heber-Midway	7.3	8.7	71.0	222.0	124.6
Deer Creek Park	7.7	8.1	115.0	192.0	159.0
Vivian Park	7.7	8.5	115.0	187.0	156.0
Deer Creek	8.1	8.3	160.0	188.0	174.0
Aspen Grove Creek	7.9	8.2	130.0	168.0	144.0
Vivian Park Creek	7.9	8.1	149.0	167.0	159.0
Highway 89	7.7	8.6	119.0	191.0	164.0
Geneva Road Crossing	7.7	8.1	142.0	176.0	159.0

passed over ice and frozen ground the solutes were less available than at other times during the year. During the spring and early summer the diversion of some of the water for irrigation purposes resulted in low stream flows occasionally being associated with small amounts of bound carbon dioxide. Flash floods occurring in the summer and fall also resulted in some slight variations in the amount of carbonate picked up from surface drainage.

BIOLOGICAL DATA

During the course of this study a total of 384 square foot samples were taken at the various stations. The total number of organisms taken was 79,298, with an average of 193 per square foot. The average volume of these organisms was 2.69 cc per square foot. The great majority (95.2 per cent) of the bottom fauna belonged to five orders of insects, the Trichoptera, Ephemeroptera, Diptera, Plecoptera, and Coleoptera. The same five orders also comprised 94.2 per cent of the total volume.

SEASONAL VARIATIONS IN PRODUCTIVITY

Since the total number of organisms taken represented many overlapping populations of individual species with different life cycles, there was considerable variation in the genera and species collected each month. With some species transforming into the adult stage and emerging during each of the four seasons, and with the precarious environment wiping out whole populations on other occasions, there was also a variation in the total numbers and volume taken each time. Even with these fluctuations in numbers, volume, and species composition, very definite seasonal trends could still be detected.

The lowest number of individuals was taken each year during the period from April to June, inclusive. In June, 1947, the average number of organisms collected per square foot was 194, these having an average volume of 1.7 cc. From April to June, 1948 there were 106 organisms per square foot, the average volume being 1.5 cc. During the same period in 1949 an average of 126 organisms per square foot was taken, with the volume averaging 1.55 cc. The small spring populations can be attributed to three causes. First and perhaps foremost, due to the high and torrential flow each spring, it was much more difficult to obtain adequate and representative samples. Second, many organisms were undoubtedly washed downstream or destroyed by the much increased flow, velocity, and molar action during the spring runoff. Third, during the late winter season and early spring, several species of stoneflies emerged as adults. In the upper portion of the stream and in the tributaries this loss was occasioned by the emergence of several different species of *Capnia* and *Nemoura*. In the lower sections *Brachyptera pacifica* emerged in large numbers from May to June each year.

Collections were made during July only in 1947. During that month the average number of organisms taken per square foot was 196, with an average volume of 1.98 cc. The principal cause of this relatively low yield in bottom fauna, was the emergence of large numbers of Ephemeroptera, Trichoptera, and Plecoptera. At the higher and intermediate altitudes large numbers of such stoneflies as *Acroneuria pacifica* and *Pteronarcella badia*, and caddis flies as *Glossosoma* spp., and *Brachycentrus occidentalis* emerged each year during June and July. Below the reservoir the emergence of *Brachycentrus occidentalis*, a stonefly, *Pteronarcys californica*, and a mayfly, *Ephemerella grandis* in June and early July greatly reduced the population left in the stream.

In August and September the populations of bottom fauna were in a constant state of flux but the standing crop was much greater both as to numbers and volume. In 1947, during these months, the average number of fauna per square foot was 423, with an average volume of 4.96 cc. In 1948 the average yield from the stream was less with only 283 organisms per square foot being collected. The average volume at that time was 3.2 cc. From these figures, it can be seen that during both years there was a decided increase in the number of immature forms during the late summer period. This increase largely consisted of the very small larvae and naiads of the species which had earlier emerged and deposited their eggs in the stream. Along with this increase in numbers, there was an increase in the size of those species which were destined to

transform to the adult stage during that period or a later date. The number of species that emerged during the period was great both as to variety and numbers. All of the major groups of aquatic insects were represented and a consideration of the species involved will be given later.

After this period of waxing and waning populations, the following six months from October, 1947, to March, 1949, saw a gradual but steady increase in the numbers of individuals represented and a very decided, marked increase in their size. The average number of individuals collected per square foot sample from October to December, 1947, was 441, with the average volume being 5.2 cc. The number increased to 619 per square foot during the period from January to March, 1948, and the volume almost doubled, being 10.26 cc. The increase in numbers was largely due to the oncoming stonefly generations of Capnia spp., Nemoura spp., and Brachyptera spp. which emerged during those months. The increase in volume was contributed by the growth of all species present.

By comparison, the corresponding six months period during the winter of 1948-49 showed a marked decrease in both numbers and volume over the autumn populations. An average of 243 organisms per square foot was taken during the months of October to November, with a reduction to 146 per square foot during February and March. The average volumes represented by these collections were 4.0 and 1.9 cc per square foot respectively. The difference in the productivity of the river during the two years can best be explained by a comparison of the weather during the two periods. The winter of 1947-48 was unusually mild with little cold weather occurring until late in December. Comparatively little ice formed over the stream at any time. The winter of 1948-49 was the most severe in 75 years, with freezing temperatures extending from early in November, 1948, to early April, 1949. A snow blanket 3 to 6 feet deep covered the ground and sections of the stream during most of that time. Air temperatures as low as -20°F caused anchor ice to form on the bottom along portions of the stream at the higher altitudes. This completely wiped out thousands of organisms.

ALTITUDINAL VARIATIONS IN PRODUCTIVITY

Nine major sampling stations were selected from a point one mile below the principal source of the river at Trial Lake to a point five miles above its outlet into Utah Lake. These stations were designated by the name of the area in which they were located or by a common reference point close to the stream (Fig. 2). Stations were selected which were most typical of the different altitudinal zones along the river and which would give the best possible picture of the stream as a whole. Six additional sampling stations were set up near the mouths of the major tributaries of the river, but these were worked on a less intensive basis.

In grading the productivity of the stream as a whole and at the various stations the standards of Hazzard (1935) were used. According to these standards a Grade 1 (exceptionally rich) stream has in excess of 50 bottom organisms per square foot, these having a volume of more than

2 cc; a Grade 2 (average) stream has in excess of 50 bottom organisms per square foot but these have a volume of only 1 to 2 cc; a Grade 3 (poor) stream has less than 50 organisms per square foot with a volume less than 1 cc. By these criteria the Provo River qualified as an exceptionally rich stream both from the standpoint of numbers and volume.

As a whole the river rated as an exceptionally productive stream but there was considerable variation in the productivity both seasonally and at different altitudes. Of 119 square foot samples taken from the upper 17 miles of the main fork of the stream, only three rated as being exceptionally rich. Seventeen samples could be classed as average, with the rest being poor in bottom fauna.

By contrast, of 100 square foot samples taken from the lower 19 miles of the river below Deer Creek Reservoir, 73 rated as being exceptionally rich, 12 as being average, and only 14 as being poor. This remarkable increase in productivity as the stream descended from 9,900 to 4,489 feet was correlated with an increase in size, volume, carbonate content, and alkalinity, and a decrease in velocity. Associated with the increase in productivity there was also a definite change in the species of organisms concerned. A description of the principal ecological features and consideration of the changes in the numbers, volume, and composition of the major groups of fauna which occurred at the different stations is presented in the following section.

Since the flow in the stream varied so greatly from season to season and from the headwaters to the mouth, measurements of both the maximum and minimum velocity and volume at each station are given in Table 1. A comparison of other important physical characteristics at each station such as average gradient, depth, width, and annual average temperature is given in Table 2.

Dissolved oxygen concentrations at all of the stations were always essentially 100 per cent, and little or no free carbon dioxide was found in the stream at any time. The differences in pH and carbonate alkalinity from the headwaters to the mouth of the stream were so significant as to justify separate consideration as given in Table 3.

The number of samples and organisms, the average volume per sample, and the food grade are given in Table 4. The taxonomic composition of the bottom fauna at each station is summarized by orders in Table 5.

1. Upper Bridge (9,500 feet).

This station was located at the first bridge crossing over the river at a point one mile below its major source, Trial Lake. The river in that area has a very rapid flow due to the steep gradient of 132 feet per mile. The stream bottom is composed largely of boulders and coarse rubble. This station and the following two were inaccessible from late November until early June each year because of the heavy snowfall which blanketed the region.

Picea engelmanni, *Abies lasiocarpa*, and *Pinus contorta murrayana* thrive in the area with some trees even lining the stream's edge. Intermixed with the trees along the river are thickets of mountain yellow willow, *Salix exigua*, and mountain meadows, supporting a variety of grasses and sedges.

No rooted aquatics occur in the stream and the growth of algae on the

Table 4. Bottom fauna per square foot sample at various stations on the Provo River, Utah, 1946-1949.

Station	No. of samples	No. of collection dates	Average organisms per sq. ft. No.	Average organisms per sq. ft. Vol. cc.	Rating Rich	Rating Ave.	Rating Poor	Ave. rating
Upper Bridge	29	16	72	0.88	1	5	10	Poor
Soapstone	29	17	68	0.86	-	6	11	Poor
North Fork	15	7	49	0.49	-	2	5	Poor
Stewart's Ranch								
Main Fork	44	25	78	0.59	2	4	19	Poor
South Fork	24	21	268	3.15	12	8	1	Rich
Lemon's Grove	34	25	421	3.19	13	5	7	Rich
Jordanelle	34	25	170	1.53	9	7	9	Ave.
Heber-Power	12	11	168	1.85	3	4	4	Ave.
Heber-Midway								
before dredging	24	20	253	2.60	8	7	5	Rich
after dredging	12	4	18	0.26	0	0	4	Poor
Deer Creek Park	27	24	472	6.33	20	3	1	Rich
Vivian Park	30	24	341	7.09	17	3	4	Rich
Deer Creek	6	3	91	0.94	-	2	1	Ave.
Aspen Grove Creek	8	7	182	2.09	3	4	-	Rich
Vivian Pk. Creek	9	7	103	1.39	3	4	-	Ave.
Highway 89	31	24	287	4.18	13	3	8	Rich

rocky bottom is poor except in the late summer and autumn, when a coat of _Zygnema_ spp., _Hydrurus foetidus_ and various diatoms may be found. Free carbon dioxide was always encountered in this section of the stream with a range of 0.5 to 2.5 ppm. The water was always nearly neutral or slightly acid with pH readings ranging from 6.8 to 7.3. The river at these higher altitudes contained distinctly soft water, the maximum carbonate content that was recorded at the Upper Bridge being only 10.0 ppm.

The average number of invertebrates taken at this station during the period of study was only 72 per square foot, these having an average volume of 0.88 cc. The river in this area and in the following 16 miles was markedly less productive than the rest of the stream.

Of the 2,095 specimens collected, 80 per cent were Ephemeroptera, Plecoptera, and Diptera. The most common species represented were the mayfly, _Baetis bicaudatus_; stoneflies, _Alloperla_ spp.; and _Acroneuria pacifica_; Diptera, _Chironomus_ spp.; caddis fly, _Arctopsyche grandis_; and beetle, _Heterlimnius quadrimaculatus_.

2. Soapstone Ranger Station (7,750 feet).

This station was located in a beautiful little mountain valley approximately 9.5 miles downstream from Trial Lake. In that area the valley is about 0.5 mile wide and the river divides in some places and slowly meanders through and around beaver ponds. The main channel through the valley has a gradient of 108 feet per mile, an average width of 32 feet, and an average depth of 1.4 feet. The stream bottom is composed largely of boulders, rubble, and coarse gravel except in the beaver ponds where a mucky bottom occurs.

In addition to the evergreens and willows found at the higher altitudes, the mountain slopes in this area are densely covered with stands of aspen, Populus tremuloides. The growth of algae on the stream bottom is more abundant than at higher elevations. Chaetophora spp. and Synedra spp. grow abundantly on all the rocks during the autumn, with Tabellaria spp., Ulothrix spp., Oscillatoria spp., and Zygnema spp. also being present.

The water at this point had only slightly different properties than at the Upper Bridge having doubled in volume, added 3°F in average temperature, and doubled in carbonate content. Other physical-chemical properties were essentially the same.

The productivity of this station was poor, with the average number of organisms taken per square foot being only 68. Stoneflies were less common and caddis flies were slightly more common. Of the 1,976 specimens collected, 68 per cent were Ephemeroptera, Trichoptera, and Diptera. The principal species found were a mayfly, Baetis bicaudatus; caddis flies, Arctopsyche grandis, and Brachycentrus spp.; midges, Chironomus spp.; stoneflies, Alloperla spp., and a beetle, Zaitzevia parvula.

3. North Fork (7,500 feet).

This fork constitutes the largest tributary of the upper Provo River, and joins the main channel about 3 miles below Soapstone Ranger Station. At its mouth it is about the same size, and has practically the same gradient and bottom composition as the upper two miles of the main stream.

The vegetation differs little from that occurring at the Ranger Station. Less algae occurs on the rocky bottom due to the increased speed of flow, but Zygnema spp. and Synedra spp. are occasionally abundant.

The drainage area of the North Fork is similar to that of the Main Fork resulting in very similar physical-chemical properties of the water. Temperatures, color, turbidity, dissolved oxygen, pH, free carbon dioxide, carbonate, and electrolyte values were essentially the same as those recorded at the Upper Bridge. Fluctuations in volume were much greater, the flow varying from 6.7 to 435.5 cubic feet per second.

Only 15 square foot samples and 726 organisms were taken at this station. The average food grade was poor. Seventy-five per cent of the organisms collected were Ephemeroptera, Trichoptera, and Diptera. The species composition was similar to that found in the upper main channel.

Table 5. Major taxonomic composition of bottom fauna by stations, Provo River, Utah, 1946-1949

Station	Total No. of organisms	Percentage in each order						
		Ephem-eroptera	Plecop-tera	Diptera	Trichop-tera	Coleop-tera	Hydra-carina	Others
Upper Bridge	2,095	30.7	27.1	22.4	7.9	2.5	-	9.4
Soapstone	1,976	34.4	12.6	15.6	18.2	5.5	13.3	0.4
North Fork	736	34.6	19.8	15.2	20.7	6.1	-	3.6
Stewart's								
Main Fork	3,439	24.7	7.0	25.6	20.5	12.2	8.5	1.5
South Fork	6,435	13.3	6.4	25.8	43.6	1.3	6.6	3.0
Lemon's Grove	14,315	7.6	5.2	13.9	62.5	7.6	1.3	1.9
Jordanelle	5,779	12.3	26.6	19.0	30.3	9.3	1.2	1.3
Heber-Power House	2,012	18.6	16.2	17.1	31.7	14.7	-	1.7
Heber-Midway Bridge	6,294	12.9	28.7	23.5	26.7	5.2	-	3.0
Deer Creek Park	10,045	39.1	2.2	22.6	24.4	-	-	11.7
Vivian Park	10,232	23.2	9.3	14.3	39.1	3.7	1.1	9.3
Deer Creek	543	63.9	2.2	12.3	17.9	1.4	-	2.3
Aspen Grove Creek	1,543	8.9	11.4	5.3	58.9	-	-	15.5
Vivian Park Creek	930	43.5	26.8	14.4	5.7	3.7	-	5.9
Highway 89	8,892	26.9	3.5	38.4	18.1	7.4	1.1	4.6

4. Stewart's Ranch (7, 100 feet).

In many respects this station was the most interesting ecologically of any studied. At Stewart's Ranch, located 17 miles southwest of Trial Lake, the Main Fork joins the South Fork and undergoes a marked change in chemical nature and productivity.

The main fork in that area has a gradient of 95 feet per mile, an average width of 35 feet, and an average depth of 1.17 feet. The stream bottom consists largely of rubble or coarse gravel. The South Fork has a gradient of 68 feet per mile, an average width of 24 feet, and an average depth of 1.0 foot. The stream bed is chiefly coarse gravel mixed with silt.

Vegetation on the mountain slopes surrounding Stewart's Ranch is like that at the upper stations. The ranch itself is located chiefly in a valley consisting of meadowland, with the tree and shrub-lined river running through it. Springs and seepage areas help to stabilize the flow of the river and support dense beds of water cress, Nasturtium officinale and water buttercups, Ranunculus spp. However, plant growth in both forks of the river itself is limited largely to algae. Oedogonium spp. and Zygnema spp. are the most common algae in both streams with Ulothrix spp. in the South Fork, and Hydrurus foetidus in the North Fork also occurring commonly.

The Main Fork of the river at Stewart's Ranch is chemically and physically similar to the three stations already described while the South Fork embodies several different characteristics. Water temperatures, color, turbidity, dissolved oxygen concentrations, and free carbon dioxide content were essentially alike in the two forks during the period of study. However, comparative temperatures meant little as a thick ice sheet covered most of the surface of the Main Fork from late December to late March each year while only the edges of the South Fork were frozen over. The average stream flow in the Main Fork was nearly four times that of the South Fork, with both volume and velocity being much more variable and violent (Table 1).

The most important chemical difference in the two streams involved the amounts of bound carbon dioxide present. No half-bound carbon dioxide appeared in either fork at any time but the carbonate content showed a vast difference. The average carbonate content of the Main Fork was 25 ppm, that of the South Fork 144 ppm, or nearly six times as great. The waters of the Main Fork were distinctly soft, those of the South Fork hard. The low carbonate content of the former is attributable to its quartzite drainage basin, while the limestone basin of the latter contributes considerable calcium carbonate to the water.

The Main Fork was similar in productivity and fauna to the upper stations already described. The South Fork was nearly four times as productive and supported a considerably different fauna. The average number of organisms collected in the Main Fork was only 78 per square foot while the South Fork produced an average of 268 specimens in a like area.

Seventy per cent of the fauna of the Main Fork belonged to the three orders Diptera, Ephemeroptera, and Trichoptera in that order of abundance. The most common species were those which were also most representative of the upper three stations. Of the organisms collected

in the South Fork Trichoptera constituted 43.6 per cent, Diptera 25.8 per cent, and Ephemeroptera only 13.3 per cent. The principal species found were a caddis fly, Brachycentrus occidentalis; Diptera, Chironomus spp., and Atherix sp.; stoneflies, Pteronarcella badia and Alloperla spp.; and mayflies, Ephemerella grandis and inermis.

5. Lemon's Grove (6,400 feet).

Lemon's Grove lies in a narrow valley ten miles southwest of Stewart's Ranch and three miles south of Kamas, Utah. The river in that area displays a slight decrease in gradient but is about one-fourth larger than the main channel at Stewart's Ranch.

Three spring-fed streams meander through the valley and empty into the Provo in the vicinity of the grove. These streams drain rich meadowland and constantly enrich the river itself. The grove consists largely of the poplars, willows, dogwood, alders, and hawthorne found at Stewart's Ranch.

An abundant growth of algae occurred in the stream bottom, particularly during the fall and winter months. This growth consisted largely of Ulothrix spp., Prasiola spp., Hydrurus foetidus, and several species of diatoms.

The mixing of the waters contributed by the two forks at Stewart's Ranch produced a noticeable physical, chemical, and biological effect by the time the stream reached Lemon's Grove. The added volume and carbonate content, and the greater alkalinity contributed by the South Fork all served to increase the productivity of the combined stream.

This section of stream could be rated as exceptionally productive, the average number of organisms taken being 421 per square foot. However, 42.1 per cent were one species, Brachycentrus occidentalis.

Of the organisms collected, 62.5 per cent were in one order, the Trichoptera, to which Brachycentrus occidentalis belongs. In addition to this species the most common forms were those contributed by the South Fork plus Baetis bicaudatus and Heterlimnius quadrimaculatus characteristic of the upper Main Fork.

6. Jordanelle (5,900 feet).

Jordanelle is located in the Heber Valley, 7.5 miles southwest of Lemon's Grove. United States Highway 40 crosses the river at that point. The area was selected as a station since the river there undergoes very great fluctuation in volume every year, due to the diversion of part of its flow for the Heber Power Plant. The sampling station was located in the section immediately below the diversion dam.

The much decreased gradient in that section, 32 feet per mile, is characteristic of the 10 miles of stream above Deer Creek Reservoir, into which the river flows. The stream bed consists largely of the boulders and coarse gravel so characteristic of the stream as a whole.

The fields on either side of the river are largely swampy meadowland. Streamside vegetation where present consists of the species of poplars, willows, alder, dogwood, and hawthorne previously listed. Dredging of the channel has resulted in steep rocky banks which restrict growth of most marginal vegetation. From late summer until early spring the rocky stream bottom is covered by a dense slimy coating of

Hydrurus foetidus. A diatom, Amphora spp. also grows abundantly at times.

Except for velocity and volume the chemical and physical properties of the stream at this station were essentially the same as at Lemon's Grove. Wide fluctuations in flow occurred with the volume varying from 1.6 to 1.062 cubic feet per second and the velocity from 0.33 to 9.2 feet per second. During periods of low flow in late summer both half-bound and bound carbon dioxide were recorded, reflecting the greater algae population and increased photosynthetic activity in the stream.

This section displayed a decided drop in productivity in comparison to Lemon's Grove. The average number of specimens collected was only 170 per square foot, these having a volume of 1.53 cc per sample. Thus the station displayed only average productivity.

Trichoptera, Plecoptera, and Diptera constituted 76 per cent of the fauna collected. The most common species represented were Brachycentrus occidentalis, Hydropsyche sp., Pteronarcella badia, Brachyptera pacifica, Chironomus sp., Baetis bicaudatus, and Heterlimnius quadrimaculatus. The large increase in numbers of Plecoptera present was due to the appearance of one species, Brachyptera pacifica.

7. Heber-Midway Bridge (5,500 feet).

This section of stream is located between the towns of Heber and Midway, Utah, 6.5 miles southwest of Jordanelle and 3 miles northeast of Deer Creek Reservoir. At the beginning of the study this section was very productive of both fish and invertebrates. Numerous deep pools and alternating riffles composed the meandering, well-shaded stream. During the summer and fall of 1947 and 1948 United States Bureau of Reclamation engineers straightened, widened, and dredged the channel in order to accommodate diversion waters for storage in the reservoir during the spring runoff. All marginal vegetation was removed for a distance of several hundred feet each side of the river, and the stream bed was increased to about twice its normal width. The stream bottom was largely coarse rubble and gravel but since dredging sand beds are becoming common.

Before the river was dredged streamside vegetation consisting of willows, dogwood, alders, and poplars lined the banks, shading the pools and keeping the water cool. Cladophora spp., Hydrurus foetidus, and various diatoms covered the stream beds providing food and shelter for herbivorous invertebrates. Now no marginal vegetation exists and the rocks in the stream look as if they had been scoured and polished.

Even before dredging this section of the stream suffered somewhat from man's abuse, but good fishing and many stream bottom organisms persisted. Water temperatures were slightly higher in late summer (71°F), and dissolved oxygen concentrations slightly lower (6.8 ppm) than at higher altitudes but the stream was still highly productive. The flow varied greatly and was reduced to dangerously low levels on occasions during late summer due to diversion of part of the stream for irrigation. At other times muddy waste water from surrounding fields was turned back into the channel increasing the turbidity from less than 7 to over 200 ppm.

Dredging provided an excellent opportunity to determine the effects

of such disturbance on the productivity of the stream. A total of 36 square foot samples was taken, 24 before and 12 after dredging. The average number after dredging was only 18, with the volume being only 0.26 cc per square foot.

Of the 6,294 organisms taken during the sixteen months before the stream bottom was disturbed 28.7 per cent were Plecoptera, 26.7 Trichoptera, and 23.5 per cent Diptera. The most common species collected were Pteronarcella badia, Brachyptera pacifica, Hesperophylax consimilis, Hydropsyche spp., Baetis spp., Ephemerella grandis, Chironomus spp., and Heterlimnius quadrimaculatus. After dredging no species was very common.

8. Deer Creek Park (5,300 feet).

This area is typical of the conditions produced in the river as a result of impoundment in Deer Creek Reservoir. The sampling station was located 0.5 mile below the reservoir's spillway and outlet pipes. The river's channel is divided in several places below the dam by islands that obstruct the flow. Many deep pools, which provide ideal resting places for the big German brown trout which inhabit the stream, are thus provided.

Two channels in the area made up the sampling station. The east branch has an average width of 60 feet and an average depth of 1.17 feet; the west branch has an average width of 28 feet and an average depth of 4.58 feet. The average gradient of the river is 61 feet per mile. The bottom consists largely of coarse rubble and gravel, with some extensive beds of fine gravel. These constitute ideal spawning beds for the trout population.

The canyon floor through which the river runs supports numerous Acer glabrum, Betula fontinalis, Quercus gambelii, Amelanchier alnifolia, Salix longifolia, Populus deltoides, Chokecherry, Prunus melanocarpa, and box elder, Acer negundo. The placid pools are deep and quiet enough to support dense beds of Potamogeton filiformis, Myriophyllum spp., and Lemna spp. The rocks in the riffles support an abundant covering of Cladophora spp., Hydrurus foetidus, and various diatoms.

Uniform, favorable conditions and a variety of habitats combined to make this section the most productive of any along the river. Water temperatures varied from 40° F to 57° F. The volume ranged between 233 and 858 cubic feet per second. Velocity ranged from 2.0 to 5.9 feet per second. The color varied from 7.0 to 20 ppm and turbidities were less than 10 ppm at all times. Dissolved oxygen concentrations were at all times approaching saturation varying from 8.1 to 11.5 ppm. Free carbon dioxide ranged from 0.0 to 2.5 ppm. The pH was within the range of 7.7 to 8.1. Methyl orange alkalinity ranged from 115 to 192 ppm.

This area was the most stable of all the stations and exceptionally productive at all times. It produced an average of 372 organisms per square foot, these having an average volume of 6.3 cc.

Of the total number of 10,945 organisms collected Ephemeroptera constituted 39.1 per cent, Trichoptera 24.4 per cent, Diptera 22.6 per cent, Diptera 22.6 per cent, and Plecoptera only 2.2 per cent. The principal species represented were Ephemerella inermis, Ephemerella grandis, Brachycentrus occidentalis, Glossosoma spp., Chironomus spp.,

Simulium spp., Arcynopteryx americana, Tubifex spp., and Glossiphonia sp.

9. Vivian Park (5,200 feet).

This station was selected, 4 miles below the preceding, because it typified mid-canyon conditions. The river is not divided into several channels in that section, but it is still characterized by deep pools and alternating runs and riffles.

The same kinds of trees and shrubs are present as occur farther upstream. However, the aquatic vegetation is restricted almost entirely to algae with Spirogyra spp., Vaucheria spp., and diatoms being common.

The physical and chemical characteristics of the stream are much the same as those at Deer Creek Park. However, the stream gradient is less steep and the flow is much more variable, due to the entrance of a tributary midway between the stations, but the environment is still stable enough to be highly productive.

An average of 341 organisms per sample was taken, giving the station an exceptionally rich rating. On a volumetric basis, 70.3 per cent consisted of only one species, Pteronarcys californica, a stonefly.

Of the total number of organisms taken, 39.1 per cent were Trichoptera, 23.2 per cent Ephemeroptera, 14.3 per cent Diptera, and 9.3 per cent Plecoptera. The most common species were the same as those listed for Deer Creek Park.

10. Highway 89 Crossing (4,489 feet).

This station is typical of lowland valley conditions but is far enough distant from the mouth of the river so as not to be affected by Utah Lake. The river at this point is about 6 miles below the mouth of Provo Canyon and 5 miles east of Utah Lake. In the summer practically all of the stream is diverted at the mouth of the canyon for irrigation purposes, leaving a mere trickle of water in the channel.

The average gradient in this section is quite similar to that in the canyon above. Coarse rubble and gravel comprise most of the stream bed with some sand piling up behind obstructions. The valley, which the river furnishes with water, is devoted largely to agriculture. Orchards and cultivated fields adjoin the stream along much of its course, being separated from the channel by only a thin strip of Salix longifolia, Populus deltoides, Acer negundo, and Carolina poplars, Populus occidentalis. Cladophora spp. and diatoms grow commonly in the riffles with Myriophyllum spp., and Ceratophyllum spp. occurring in the quieter pools. Polygonum spp., grasses, and various weeds grow along the margins in many places.

Stream temperatures at this station were consistently higher than at higher altitudes, the average being 55.3° F. The flow varied considerably, the volume reflecting the irrigation needs of the valley. Dissolved oxygen concentrations were never lower than 7.6 ppm and were always near saturation. The water was always slightly alkaline and averaged 164 ppm dissolved carbonates.

Despite the wide fluctuations in water level which occurred, this station was exceptionally productive of botton invertebrates. The average

number of organisms taken per square foot was 287 representing a volume of 4.18 cc per sample. Diptera constituted 38.4 per cent of the organisms collected, Ephemeroptera 26.9 per cent, Trichoptera 18.1 per cent, and miscellaneous orders 16.6 per cent. The most common species were also common at Deer Creek Park and Vivian Park but midge and blackfly larvae and pupae were far more abundant here than at any other station.

11. Tributary Stations.

In addition to the major sampling stations already considered, three tributaries entering the river in Provo Canyon were studied but on a less regular basis. The three, Deer Creek, Aspen Grove Creek, and Vivian Park Creek, displayed very similar physical and chemical properties throughout the period of study. All three are clear, cold, rapid mountain streams with considerable variation in flow, an abundance of dissolved oxygen at all times, no free carbon dioxide at any time, a decidedly alkaline pH, and hard water with a high carbonate content. Aspen Grove Creek was the most productive, yielding an average of 182 organisms per square foot, while the other two were only average in productivity. The fauna in all three streams was similar but the proportions were quite different (Table 5). Trichoptera and Plecoptera comprised 70 per cent of the fauna in Aspen Grove Creek; Ephemeroptera and Trichoptera (80 per cent) were most common in Deer Creek; while Ephemeroptera and Plecoptera constituted 70 per cent of the fauna in Vivian Park Creek.

SUMMARY

The Provo River, Utah, a typical mountain stream of the Intermountain Region, was studied between September 15, 1946 and May 30, 1949 with particular reference to the ecological factors which were correlated with the rich productivity of invertebrate animals in the river. Nine major sampling stations and six supplementary ones were selected in typical average sections of the stream. A series of 267 physical and chemical determinations were made at these stations. A total of 384 square foot samples was taken during the period of study. The number of organisms collected averaged 193 per square foot, with an average volume of 2.69 cc per square foot. By the standards of Hazzard (1935) the Provo River rated as an exceptionally rich stream.

Since the total number of organisms taken represented many overlapping populations of individual species, representing many different but characteristic life cycles, there was considerable variation in the composition of the samples obtained. Even with these variations, very definite seasonal and altitudinal trends were detected. The period from April to June each year yielded the smallest number of organisms per square foot sample. The number and volume of organisms per sample were greatest from December to March. The winter of 1948-49, however, was particularly severe and the populations declined during this period.

There was a gradual increase in the productivity of the stream as it descended from its headwaters to Utah Lake. This increase reached its

peak in the five mile section below Deer Creek Reservoir, a section in which the environment was far more stable than elsewhere along the river. Of 119 square foot samples taken from the upper 17 miles of the main fork of the stream, only 3 rated as being exceptionally rich. Of 100 samples taken from the lower 19 miles of the stream, 73 rated as being exceptionally productive. The productivity and composition of the fauna at several of the stations at the lower altitudes were affected by man's activities but nature displayed remarkable powers of recovery from the unfavorable conditions which man imposed. Of all the factors considered, greater uniformity in flow, higher and more uniform temperatures, a more alkaline pH, and a higher carbonate content were most responsible for the increase of productivity in the lower sections of the stream.

The great majority (95.2 per cent) of the bottom fauna belonged to five orders of insects, the Trichoptera, Ephemeroptera, Diptera, Plecoptera, and Coleoptera. The same five orders also comprised 94.2 per cent of the total volume. The most abundant and widely distributed organism encountered was a caddis fly, Brachycentrus occidentalis. This species constituted 12.3 per cent of all the organisms collected. A stonefly, Pteronarcys californica, comprised 22.4 per cent of the total volume, the most abundant species other than these two were three mayflies, Baetis tricaudatus, Ephemerella inermis and E. grandis; a stonefly, Pteronarcella badia; and a beetle, Heterlimnius quadrimaculatus.

Throughout the course of the study the variations in color, turbidity, dissolved oxygen, and free carbon dioxide content at all of the stations and during the different seasons were so slight that those factors could not be considered significant in explaining the differences in productivity at various altitudes along the stream. The differences can best be explained on the basis of changes that occurred in the nature of the stream bed, the velocity, the volume, the temperature, the pH, and the carbonate content as the river descended from its lofty origin to the valley floor.

A stream bed of large boulders or rubble, great variability in velocity, volume, and temperature, neutral or slightly acid water, and low carbonate content were correlated with low productivity in the river at the higher altitudes. A stream bed of coarse gravel and silt, greater uniformity in velocity, volume, and temperature, uniformly alkaline water (pH 7.7 or above), and high carbonate content were associated with high productivity in the river at the lower elevations.

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